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DECEMBER, 1949

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Membership: total (Nov. 10, 1946), 627. *Eligibility* (*Const., Art. 14, Sec. 1*).—"Any person who has conducted, and published an account of, a meritorious original investigation in dental science, or in any of the sciences contributory to stomatology, shall be eligible to membership in this Association, providing such person conforms to the recognized standards of professional ethics." *Election.*—New members may be elected *only at general meetings of the Association.* Membership in a section is secondary to membership in the Association as a whole. Removal of members from one geographic center to another does not affect their membership in the Association.

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Lazansky, Robinson, and Rodofsky (p. 533) investigated the occurrence of bacteremias after the extraction of teeth and found it to be much less (10 per cent) than that found by most other authors (40 per cent). They do not attempt to explain the difference between their results in 221 operations and those of the other investigators in 1,078 operations. The bacteremias were influenced by the age of the patient, the amount of trauma, and the number of teeth extracted. The authors agree, however, that even if teeth are extracted singly from patients with rheumatic fever or congenital valvular disease premedication is indicated.

Too often new therapeutic agents are conceived on the basis of loose thinking, marketed and, later, subjected to clinical trial. Rothner, Cobe, Rosenthal, and Bailin (p. 544) discuss the results of clinical trial of a carboxymethyl cellulose base containing penicillin. The drug is applied to the gingiva in acute infections amenable to penicillin therapy. The results reported indicate a marked antibacterial effect, but, as the authors state, drug therapy does not reduce the need for removal of irritating factors.

Sognnaes (p. 549) using a special technic with great care has obtained specimens of enamel which he has examined under the binocular scope during microdissection and under higher power. When technics such as this are developed, great caution must be used to prevent misinterpretations. Consultation between oral histologists can be as valuable as that between clinicians. For example, care must be taken in explaining the Schreger's bands, optically, in view of the demonstration of variations in the Grenz-ray pattern, i.e., density (Applebaum: *J. D. Res.* **22**: 7, 1943).

Sognnaes (p. 558) continuing the study of his details of the organic structure of enamel has observed and photographed thickenings of the neonatal lines and striae of Retzius. He suggests that this is contrary to the common description of these lines as areas of less calcified components of the enamel rod. While technically there is a difference, by and large, an area of widened organic prism sheaths is also an area of less calcified matter, i.e., enamel rod substance. The darker component, running at right angles to the sheaths (Fig. 2), might also be interpreted as part of the increased organic material of the rod itself.

It is encouraging to see a research report from the University of the Philippines. Villa (p. 565) has observed a membranous structure at the dentino-enamel junction of two decalcified teeth which is interpreted as the dentino-enamel cuticle. The observation is interesting but Villa's tendency to generalize about its relationship to the carious process on the basis of its appearance in a single carious lesion is highly dangerous. Too much dental histology and pathology has been based on studies of one or two sections. While it is recognized that obtaining good specimens of decalcified carious teeth is difficult, this cannot serve as an excuse for generalization from isolated examples.

Phillips and Swartz (p. 569) found a great variation in the mercury content of old amalgam restorations removed from extracted teeth. There was no greater content of mercury in multiple surface than in one surface restorations. The extremes of 28.6 per cent and 60 per cent mercury content indicate the magnitude of variation found.

Cresson (p. 573) has critically examined the A.D.A. Specification #11 for hydrocolloid impression materials and the Medical Department Tentative Specification No. MD-1285 for alginate impression materials and concludes that they do not give a true measure of the value of such materials. He has devised and outlined new tests for evaluating impression materials. It is well to review and, at times, revise specifications of dental materials. The Materials Group of the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH is in an ideal position to discuss and suggest such changes when they are advisable.

Studying the hydrogen-ion concentration of the saliva, Anderson (p. 583) confirmed the findings of others that there is a "remarkable constancy" of salivary pH. A statistically significant fall in $[H^+]$ between 3:00 P.M. and 3:45 P.M. may be related to the alkaline tide. Ingestion of alkali and acid were reflected in the saliva but the author states that the role of the salivary glands in acid-base regulation would be of very little importance in comparison with that of the kidneys.

Rae and Clegg (p. 589) attempted to correlate buffering capacity, viscosity, and salivary lactobacillus counts of 70 individuals. It is interesting to see such correlated studies for certainly they are more acceptable than assumptions based on mathematical exercises with different sets of data. The authors failed to find correlations suggested by other investigators. They demonstrated a marked constancy of viscosity, tested by laboratory methods, between salivas of different individuals. This is in contrast to rule of thumb studies made by observing the "stringing" of saliva from the mouth.

Hadjimarkos and Storvick (p. 594) found occurrence of caries less in a group of institutionalized children on an "optimal" diet and restricted refined carbohydrate intake than in other Oregon children. The authors believe both factors, refined carbohydrate intake and optimal diet, to be factors in reducing caries in these children, but do not undertake to explain how either factor may operate. It may be that reduction in the number of times confections are consumed may be important. If each time refined sugar is taken into the mouth one-half hour of acid activity takes place under the bacterial plaques, then three meals a day with carbohydrates would result in 1½ hours of decalcification. The consumption of confections at 3 times between meals per day, common practice in noninstitutionalized children, would double the time for decalcification.

Russell (p. 600) investigated three groups of children exposed to fluorinated water for 18 months, one group exposed before eruption of teeth, one exposed before, during, and after eruption of permanent teeth, and one continuous-resident group. He hypothesized, after consideration of the data, that fluorine may be incorporated into enamel before or after eruption and inhibit caries, that this protective action tends to be lost after fluoride exposure is discontinued, and that periodic or continuous renewal of fluorides is necessary for maximum protection. Russell's data and his hypotheses must be considered if artificial fluorination data is to be properly interpreted and, also, in their possible relationship to repeated topical fluorinizations.

Zipkin and McClure (p. 613) have carefully adapted analytical methods for determination of citric acid in saliva. Using these methods they have determined the salivary citrate acid content of 60 individuals with erosion and 61 without erosion. A statistical relationship between erosion and salivary citrate was established. It is unusual that erosion was observed in 27 per cent of 83 patients selected at random. Shulman and Robinson had found only 2 per cent in 1,345 college freshmen. There is a tendency to group dentifrice abrasion with erosion, but commercially available dentifrices are not abrasive enough to cut through enamel under normal circumstances. Dentifrice abrasion can be differentiated because it originates in exposed dentin on the roots of teeth. Perhaps, if those individuals with pure abrasion had been removed from Zipkin and McClure's group the citrate-erosion relationship might have been more striking. The subject of erosion-salivary citrate relationship should be further investigated.

The Proceedings of the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH (p. 627) reflect continued growth of the ASSOCIATION and of dental research. President Blayney's address reviews the history of the scientific growth of the ASSOCIATION; the one hundred thirty-one abstracts reflect it.

Among the new books we have recently received are: Paul Boyle's revision of Kronfeld's **Histopathology of the Teeth** (Philadelphia, 1949, Lea & Febiger), a welcome revision of an old favorite; J. M. Dyce's **Photographic Aids to Clinical Dental Practice** (New York, 1948, Staples Press Ltd.) which is a beautifully illustrated book that should answer a need of the many dentists who are employing the camera as an aid to practice and teaching; James T. Ginn's **Review of Dentistry** (St. Louis, 1949, The C. V. Mosby Company), a question and answer book which will probably find its chief use in preparing for State Board examinations; Henry Goldman's second edition of **Periodontia** (St. Louis, 1949, The C. V. Mosby Company), a real revision which appears to offer a well-balanced discussion; Willy Geier's **Die Problamatik der Partiellen Prothese** (Konstanz A. B., Postfach 280 Verlag Zahnaerzliche Welt G. M. B. H:1948) is the first of new books from Germany that we have seen in the postwar era; and R. A. Willis' **Pathology of Tumors** (St. Louis, 1948, The C. V. Mosby Company), a thorough discussion of neoplasms with definite indications through nomenclature and theories of its background in "British" pathology.

H. B. G. R.

FACTORS INFLUENCING THE INCIDENCE OF BACTEREMIAS FOLLOWING SURGICAL PROCEDURES IN THE ORAL CAVITY

JOSEPH P. LAZANSKY, M.D., D.M.D., LEONARD ROBINSON, A.B., D.M.D., AND LILLIAN RODOFSKY

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BACTERIA have been isolated from the blood stream for varying periods of time following such surgical procedures as tonsillectomy,[1, 2, 3] orthopedic surgery,[4] manipulations of the genitourinary tract,[5, 6] and ear operations.[7, 8] There are also reports of transient bacteremia associated with gingival massage,[9] mastication,[10, 11] rocking[12] and extraction of teeth.[13-21] These bacteremias are said to be of varying duration, ranging from five minutes to several hours after operation.[18] They have been attributed to dissemination of organisms from: (a) areas of infection at the apices of teeth,[15] (b) the submucous tissue as a result of injection of the anesthetics,[10, 22, 23] and (c) the gingival sulcus.[14] The incidence of bacteremia has been correlated with the degree of periodontal disease,[12, 13] the type anesthetic employed,[12, 14] and the amount of trauma involved.[12, 13] The effects of sulfonamides and penicillin in affecting both the incidence and duration of bacteremia have been reported.[19, 21] Clinical histories are available that both support and refute a relationship between dental surgical procedures and the development of subacute bacterial endocarditis.[24-28]

Studies on the rate of clearance of bacteria from blood,[29] together with the rate of blood circulation,[30] indicate that bacteria are quickly removed from the blood stream. Because evidence concerning the factors responsible for the occurrence and duration of bacteremias of dental origin is inconclusive, it was felt that further study of these factors was warranted so that prophylactic measures could be more properly directed and more effectively used.

METHODS

Patients.—One hundred twenty-five ambulatory patients presenting themselves for treatment at the Oral Surgery Clinic of the dental school were studied. Of these, forty-six consented to participate two or more times, so that a total of 221 operations were performed.

Blood Samples.—Blood was withdrawn from one of the veins of the antecubital fossa after the overlying skin had been surgically prepared with tincture of iodine followed by alcohol. In the initial 161 cases, three 10 c.c. blood samples were taken. A preoperative sample (sample A) was withdrawn 15 to 30 minutes prior to operation. A second sample (sample B) was taken within the first minute following completion of the operation, while the third blood specimen was withdrawn after ten minutes had elapsed. Since in the first 73 cases no bacteria were recoverable ten minutes after operation, the ten-minute interval was reduced to five minutes for the remainder of the study.

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The data represent part of the material to be submitted as a partial requirement for the degree of Master of Science, Tufts College.

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Because the preoperative blood sample in the initial 161 cases yielded no bacteria, it was discontinued as a routine procedure. The amount of Sample B (taken within the first minute postoperatively) was increased to 20 c.c.

Bacteriological Procedures.—Blood samples were transferred to flasks containing 100 c.c. tryptic digest broth (enriched by 0.1 per cent dextrose). In the early phases of the study, the flasks were subcultured as soon as any cloudiness appeared. It was found, however, that some hemolysis took place after seventy-two hours, and that the contents of most of the flasks became cloudy. Addition of sodium citrate in an attempt to reduce hemolysis was accompanied by increased contamination. For this reason, it was discontinued, and the procedure of routinely subculturing the flasks on the seventh and fourteenth days was adopted.

Streptococci were identified on the basis of (a) type growth on blood agar plates and (b) sugar fermentations. The classification of streptococci is that of Frost and Englebrecht (Table I).

At the beginning of the study, pour plates consisting of 1 c.c. of the patient's blood, tryptic digest agar, and 5 per cent horse blood were made in an attempt to evaluate bacteremias quantitatively. Since the only organisms which grew on the plates were considered contaminants, the procedure was discontinued. Socket cultures were also taken early in the study, but were discontinued because no correlation was observed between organisms of the socket culture and organisms responsible for bacteremia.

When it became apparent that the incidence of positive blood cultures was much less than that already reported, a study was made of the effectiveness of tryptic digest broth (containing 0.1 per cent dextrose) as a culture medium. The growth-promoting ability of the medium was compared with (a) tryptic digest broth containing 1 per cent dextrose, and (b) peptone broth. A recently isolated *Str. viridans* culture incubated less than twenty-four hours was diluted successively, resulting in a series of dilutions ranging from 10^{-1} to 10^{-11}. Each dilution was cultured in triplicate in each of the three media, and seeded on two pour plates, one of which contained human blood serum, and the other contained horse blood serum.

It was found that the test organisms appeared in all three media in all dilutions up to 10^{-7} inclusive. No growth occurred in any of the media at greater dilutions. The organisms grew equally well in all three media. Colony counts on the pour plates were essentially the same for human and horse blood serum. It was therefore concluded that tryptic digest broth containing 0.1 per cent dextrose compared favorably with other media frequently employed in bacteriological studies.

Surgical Procedures.—The 221 operations are divisible into four groups. In Group I, consisting of 34 patients, only single teeth were extracted. Group II was composed of 22 patients. In these, single teeth were initially extracted, but following withdrawal of the second postoperative blood sample (Sample C), an

additional two or more teeth were extracted, and another blood sample was taken immediately following the extractions. In Group III, two or more teeth were extracted initially in 70 patients. Group IV consisted of 72 cases in whom periodontal scalings were performed. In the majority of cases, scaling was carried out on the four mandibular incisors and for a period of ten minutes. In one patient, an alveolectomy was performed, but no positive blood cultures were obtained.

TABLE I
CLASSIFICATION OF STREPTOCOCCI (AFTER FROST AND ENGLEBRECHT)

ORGANISM	SUGAR FERMENTATION								TYPE COLONY
	LACTOSE	MANNITE	SALICIN	SORBITOL	RAFFINOSE	INULIN	SACCHAROSE	TREHALOSE	
A. Nonhemolytic Streptococci									
Str. inulinaceus	+	+	+	–	+	+	+		alpha
Str. faecalis (Enterococcus)	+	+	+	+	+	–	+		alpha
Str. distendens	+	+	+	+	–	+	+		alpha
Str. liquefaciens	+	+	+	+	+	–	+		
Str. lactis	+	+	+	–	–	–	–		alpha
Str. non-hemolyticus I	+	+	–		–	–			
Str. bovis	+	–	+	–	+	+	+		alpha
Str. cardio-arthritidis	+	–	+		+	+	+		gamma
Str. bovinus	+	–	+	–	+	–	+		alpha
Str. anhaemolyticus	+	–	+		+	–	+		gamma
Str. pneumosimilis	+	–	+	–	–	+	+		alpha
Str. agalactiae	+	–	+	–	–	–	+		alpha
Str. mitis	+	–	+	–	–	–	+		alpha
Str. cremoris	+	–	+	–	–	–	–		gamma
Str. salivarius	+	–	–	–	+	–	+		alpha
Str. morbilli	+	–	–	–	+	–	+		alpha
Str. thermophilus	+	–	–	–	–	–	+		gamma
Str. non-hemolyticus II	–	+	+			–			
Str. non-hemolyticus III	–	+	–			–	+		
Str. equarius	–	–	+		–	+			alpha
Str. equinus	–	–	+		–	–	+		alpha
Str. ignarvus	–	–	–		+	+	+		gamma
B. Hemolytic Streptococci									
Str. infrequens	+	+	+	+	–	–	–	+	
Str. hemolyticus I	+	+	–				+		
Str. zooepidemicus	+	–	+	+	–	–	+	–	
Str. pyogenes	+	–	+	–	–	–	+	+	
Str. epidemicus	+	–	+	–	–	–	+	+	
Str. erysipelatos	+	–	+	–				+	
Str. scarlatinae	+	–	+	–				+	
Str. asalignus	+	–	–	–	–	–	+	+	
Str. aginosus	+	–	–	–	+	+	+	–	
Str. alactosus	–	+	+	–	–	–	+	+	
Str. hemolyticus III	–	+	–						
Str. equi	–	–	+	–	–			–	
Str. equisimilis	–	–	+		+			–	
Str. subacidus	–	–	–			–	+		
Str. mastitidis	+	–	+	–	–	–	+	+	

Prior to operation, a pertinent medical history was elicited from each patient. The vitality of each tooth involved in the operation was tested by means of a Burton Vitalometer. Tooth mobility, gingival inflammation, pocket formation, presence or absence of free pus, and the amount of calculus were noted. Full mouth roentgenographs of each patient were available and were studied for the presence or absence of apical areas of radiolucence, root canal fillings, and alveolar bone loss. The amount of manipulation required and the duration of the operation were recorded.

RESULTS

Frequency and Distribution of Positive Blood Cultures and the Type Organism Involved.—Transient bacteremias were detected 22 times in 221 operations, an over-all incidence of 10 per cent. The organisms were alpha hemolytic streptococci except for one case in which the organism isolated was a nonhemolytic (gamma) streptococcus. No mixed cultures were obtained. (Table II.)

TABLE II
FREQUENCY AND DISTRIBUTION OF POSITIVE BLOOD CULTURES

GROUP	NO. OF CASES	NO. OF POSITIVE	BLOOD CULTURES POSITIVE	
			BLOOD SAMPLE TAKEN WITHIN ONE MINUTE AFTER OPERATION	BLOOD SAMPLE TAKEN FIVE MINUTES AFTER OPERATION
I Single extractions	34	1	1 *Str. inulinaceus*	
II (a) Single	22 (56)			
(b) Multiple extraction following single extraction	22 (92)	4	4 *Str. salivarius* (2) *Str. inulinaceus* (1) *Str. equinus* (1)	
III Multiple extractions	70	12	10 *Str. anhaemolyticus* (1) *Str. pneumosinilis* (1) *Str. bovis* (2) *Str. inulinaceus* (3) *Str. non-hemolyticus* (1) *Str. bovinus* (1)	3* *Str. bovis* (2) *Str. inulinaceus* (1)
IV Periodontal scalings	72	5	3 *Str. inulinaceus* (1) *Str. bovis* (1) Nonhemolytic streptococcus (Gamma) (1)	2† *Str. bovis* (2)

*These three samples were withdrawn five minutes after operation. In one case, both one-minute and five-minute samples were positive. In the other two, the previous one-minute samples were negative.

†In these the preceding one-minute samples were negative.

In 56 single extractions, one positive blood culture was obtained (2 per cent). In 92 multiple extractions, positive blood samples occurred in 16 of the cases (17 per cent). In computing results, the findings on the 22 patients in Group III were included in both single and multiple extractions. In 72 periodontal scalings, positive blood samples resulted in 5 cases, an incidence of 7 per cent.

In only five cases were bacteremias observed in the five-minute samples. In four of these, the preceding one-minute samples were negative. No bacteria were recovered in the ten-minute samples.

A complete summary of the data is presented in Table IV. The results indicate that the incidence of detectable bacteremia is not influenced by sex, tooth vitality, tooth mobility, periodontal disease, the location of the tooth or teeth, or by the presence of apical infections. Positive history of foci of infection other than dental did not appear to predispose to bacteremia.

Three factors do appear to exert definite effects on the incidence of bacteremia resulting from dental procedures. These are age, number of teeth extracted, and the amount of operative trauma.

Age.—The data indicated that bacteremias tended to occur most frequently in the younger and older age groups. The influence of age was therefore studied by grouping the patients less than 30 years of age with those 60 years or older, and comparing this composite group with the age group 30 to 59 inclusive. A significantly greater incidence of transient bacteremia was noted in this composite group of extremes. This difference was observed when the scaling, single extraction, and multiple extraction groups were considered together, when single and multiple extractions were considered together, or when multiple extractions were considered alone. (Chi-square values* in all three instances ranged from 4.63 to 9.41.)

Number of Teeth Involved.—The incidence of bacteremia is significantly greater when two or more teeth are extracted than when single teeth are extracted (2 per cent versus 17 per cent, and a Chi-square value of 8.34). When more than one tooth is extracted, the actual number of teeth does not appear to be a significant factor.

Trauma.—The trauma incurred during each operative procedure was evaluated at time of operation as slight, moderate, or extensive. In only eight cases was it considered extensive. For this reason, moderate and extensive were considered together. The role of trauma was studied by comparing the incidence of detectable bacteremias in cases where trauma was slight with that in the cases where trauma was moderate or extensive. The incidence of detectable bacteremia was greater in operations accompanied by moderate or extensive trauma (8 per cent versus 17 per cent). This difference was noted when single and multiple extractions were considered together, and when multiple extractions were considered alone. (Chi-square values in both instances were 9.2 and 5.7, respectively.)

Incidence of Bacteremias in Patients Employed Two or More Times.—In the 46 patients utilized two or more times in the study, only two yielded positive blood samples twice. There does not appear to be a predisposition toward the development of bacteremia by certain individuals.

Contamination.—In eighteen instances, the blood samples were found to contain organisms other than streptococci. Unclassified diphtheroids were isolated in ten, *Staphylococcus albus* in seven, and a gram-positive spore-forming bacillus in one. In ten cases, the organisms were detected only after a fourteen-

*Chi-square values were obtained by using the fourfold table.[21]

day interval; in two the organisms appeared after seven days, while in five the organisms were isolated before seven days had elapsed. Diphtheroids and *Staph. albus* tend to grow rather quickly, appearing in media after an interval of twenty-four to seventy-two hours. Because of this fact, and the fact that in most instances the detection of these organisms coincided with opening of flasks for routine seven-day and fourteen-day subcultures, it was felt that the positive blood cultures resulted from contamination and were not due to blood stream invasion.

DISCUSSION

The fact that no positive blood cultures were obtained preoperatively in our initial 161 cases is in general consistent with the findings of other workers. In a total of 1,078 instances of tonsillectomy and tooth extraction where preoperative samples were taken, 31 positive blood cultures were obtained preoperatively. It is of interest that 19 of these occurred in two studies carried out by Okell and Elliot,[13] and by Elliot alone.[12] In no other study were there more than three preoperative positives. Eight of the 31 reported were considered by the investigators concerned to be the result of contamination. The remainder provides an incidence of preoperative bacteremias amounting to merely 2 per cent.

In several respects, however, the results obtained here differ significantly from those previously reported. Points of difference are (1) the incidence and duration of bacteremia following dental procedures, and (2) the influences of the number of teeth extracted and of gingival inflammation.

Incidence.—A summary of results obtained by other investigators discloses that 482 bacteremias occurred in 1,187 cases involving extraction of teeth, gingival massage, or mastication (Table III). This is an incidence of 40 per cent. Two investigations alone, that of Okell and Elliot,[13] and that of Murray and Moosnick,[11] both together involving 474 patients, accounted for 269 of the 482 bacteremias. Even if these two are not included in the totals, the incidence reported is 30 per cent. This is still significantly higher than the 10 per cent observed in our study.

Duration.—In only five instances in our study was a bacteremia detected five minutes after instrumentation, and in no case was it found ten minutes after. This is in contrast to reports of other investigations, in which 153 positive blood cultures were obtained within five minutes (usually two to three minutes) following extraction, and in which 56 positive blood cultures were obtained ten minutes after operation. In Okell and Elliot's study,[13] all 84 positive postoperative blood cultures occurred in samples taken from two to five minutes after extraction. In the study by Burket and Burn,[14] 44 positive blood cultures resulted in the ten-minute blood samples, the only postoperative samples taken.

There were four cases in our study in which five-minute samples were positive, while the preceding postoperative samples were negative. Perhaps this can be explained on the assumption that the bacteria are not diffusely distributed in the blood; there is, therefore, a possibility that a certain percentage of samples of blood from cases with bacteremia might not contain any bacteria or enough to give a positive culture.

TABLE III
SUMMARY OF RESULTS REPORTED BY OTHER INVESTIGATORS

	INVESTIGATOR	NO. OF CASES	NO. OF POSITIVE POSTOPERATIVE CULTURES
Single extractions only	Okell and Elliot[13]	10	1
	Burket and Burn[14]	188	44
	Elliot[12] (Rocking of single teeth)	41	23
	Glaser, Dankner, Mathes, and Harford[21]	16 (control group)	10
	Marseille[15]	100	42
		Total 355	Total 120
Multiple extractions only	Okell and Elliot[13]	128	83
	Bender and Pressman[20]	30 (control group)	25
	Glaser, Dankner, Mathes, and Harford[21]	24 (control group)	17
		Total 182	Total 125
One or more extractions	Palmer and Kempf[17]	82	13
	Northrop and Crowley[19]	97 (control)	16
	Hopkins[16]	108	18
		Total 287	Total 47
Chewing, gingival massage, etc.	Richards[9]	17	3
	Round, Kirkpatrick, and Hails[10]	10	2
	Murray and Moosnick[11]	336	185
		Total 363	Total 190
Total		1187	482

Number of Teeth Extracted.—Our findings of one bacteremia in 56 cases of single extraction (2 per cent) differs from the composite results of other studies where 120 positive blood cultures were obtained in 355 single extractions (33 per cent). Likewise, the occurrence of 16 bacteremias in 92 instances (17 per cent) where two or more teeth were extracted is less than the 125 positives reported in 182 patients (68 per cent). In the study by Glaser, Dankner, Mathes and Harford,[21] the incidence of bacteremias among 38 cases following multiple extractions is significantly higher than that among 42 cases of single extractions (Chi-square value = 7.5). The composite results of other studies are of a higher magnitude than was obtained in the present investigation, but they still reflect our observation that multiple extractions are more conducive to the development of transient bacteremia than are single extractions. Four of the 22 patients in Group III developed bacteremia following multiple extractions, whereas in none were positive blood cultures obtained after single extractions. This is consistent with the observation just mentioned even though it is not statistically significant.

Periodontal Disease.—In this study, periodontal disease did not predispose to bacteremia. Gingival inflammation, or pocket formation, or the presence of purulent exudate about the necks of the teeth were considered as evidences of periodontal disease. There are conflicting reports in the literature as to the importance of periodontal disease in this respect. Okell and Elliot[13] reported that the incidence of bacteremia among extraction cases when moderate or marked gingival disease was present was significantly greater than when it was absent. Elliot,[12] in studying the effects of rocking teeth, found the incidence

TABLE IV

SUMMARY OF DATA: BACTEREMIA FOLLOWING PERIODONTAL SCALINGS AND EXTRACTIONS OF TEETH

		SINGLE EXTRACTION CASES (56)				MULTIPLE EXTRACTION CASES (92)				PERIODONTAL SCALING CASES (72)			
			No.	+	-		No.	+	-		No.	+	-
Sex	Male		28	1	27		36	7	29		41	4	37
	Female		28	0	28		56	9	47		31	1	30
	Total		56	1	55		92	16	76		72	5	67
Age	10 to 19		1	0	0		2	0	2		0	0	0
	20 to 29		14	0	14		19	7	12		18	0	18
	30 to 39		15	0	15		24	2	22		22	2	20
	40 to 49		9	0	9		26	3	23		16	2	14
	50 to 59		10	0	10		14	1	13		6	0	6
	60 to 69		7	1	6		7	3	4		6	1	5
Season	Jan., Feb., March		49	1	48		21	3	18		23	2	21
	April, May, June		6	0	6		24	7	17		31	3	28
	July, Aug., Sept.		0	0	0		16	6	10		17	0	17
	Oct., Nov., Dec.		1	0	1		31	0	31		1	0	1
Teeth involved	Maxilla		26	0	26		53	9	44		13	1	12
		I*	5	0	5	A†	6	0	6	A	2	1	1
		C	3	0	3	P	22	3	19	P	1	0	1
		P	8	0	8	B	25	6	19	B	10	0	10
		M	10	0	10								
	Mandible		30	1	29		34	6	28		57	3	54
		I	6	0	6	A	6	1	5	A	41	2	39
		C	2	0	2	P	9	2	7	P	2	0	2
		P	7	1	6	B	19	3	16	B	14	1	13
		M	15	0	15								
	Both maxilla and mandible						5	1	4		2	1	1
						A	0	0	0	A	0	0	0
						P	3	1	2	P	1	0	1
						B	2	0	2	B	1	1	0
Tooth vitality	Vital		45	1	44		50	8	42		62	5	57
	Nonvital		11	0	11		12	3	9		0	0	0
	Both vital and nonvital						30	5	25		10	0	10
Root canal filling	Present		2	0	2		0	0	0		0	0	0
	Absent		54	1	53		89	16	73		71	5	66
	Both						3	0	3		1	0	1
Tooth mobility	None		25	1	24		44	7	37		42	4	38
	Slight		18	0	18		25	6	19		16	1	15
	Moderate		11	0	11		12	1	11		10	0	10
	Marked		2	0	2		11	2	9		4	0	4
Gingival inflammation	Present		25	0	25		53	11	42		50	1	49
	Absent		29	1	28		39	5	34		21	4	17
Pocket formation	None		11	0	11		16	4	12		30	1	29
	Slight		15	1	14		30	2	28		14	2	12
	Moderate		20	0	20		31	7	24		22	2	20
	Marked		9	0	9		15	3	12		6	0	6
Free pus	Present		14	0	14		42	8	34		33	1	32
	Absent		42	1	41		50	8	42		38	4	34

TABLE IV—CONT'D

		SINGLE EXTRACTION CASES (56)			MULTIPLE EXTRACTION CASES (92)			PERIODONTAL SCALING CASES (72)		
		NO.	+	−	NO.	+	−	NO.	+	−
Calculus	Absent	23	1	22	45	6	39	3	1	2
	Slight	6	0	6	14	1	13	32	2	30
	Moderate	2	0	2	5	1	4	12	2	10
	Marked	2	0	2	0	0	0	19	0	19
Manipulation	Slight	51	1	50	68	8	60	36	2	34
	Moderate	3	0	3	18	5	13	36	3	33
	Marked	2	0	2	6	3	3	0	0	0
X-ray findings	Apical areas: Present	12	0	12	41	8	33	5	0	5
	Absent	44	1	43	51	8	43	61	5	56
	Bone loss: None							14	2	12
	Slight	Not observed			Not observed			27	2	25
	Moderate							22	1	21
	Marked							3	0	3
Local anesthesia	Infiltration	30	0	30	56	9	47			
	Block with infiltration	26	1	25	34	7	27	Not employed		
History of other foci of infection	Present	7	0	7	3	1	2	1	0	1
	Absent	49	1	48	89	15	74	71	5	66

*I = incisor, C = cuspid, P = premolar, M = molar.
†A = anteriors, P = posteriors, B = both.

of positive blood cultures to be greater in the presence of gingival disease. In the study by Burket and Burn,[14] a suspension of *Serratia marcescens* was painted around the necks of the teeth to be extracted in 90 patients. In 18 cases this organism was recovered from the blood stream, while in 15 others, organisms other than *S. marcescens* were detected. It appears, therefore, that although the gingival crevice can be a source of entry for organisms, there are probably other sites of equal or greater importance. An analysis of the results of Glaser, Dankner, Mathes, and Harford[21] reveals that no significant difference existed between the normal and abnormal gingival cases.

Conflicting claims exist concerning the roles of conduction and infiltration types of anesthesia in the production of bacteremia. It has been suggested that when infiltration anesthesia is employed, the vasoconstrictor present in the anesthetic tends to prevent entry of bacteria from the operative site. Others hold that deposition of anesthetic at the site of operation is traumatic and therefore increases the possibility of bacteria gaining entrance into the blood. It is therefore suggested that fewer bacteremias occur with conduction than with infiltration anesthesia. In the present study, conduction anesthesia was always supplemented by terminal infiltration, and it is not possible to evaluate the role played by either one.

Clinical Implications.—The incidence of bacteremia following single extractions is negligible. For this reason it appears that the procedures we employed

do not occasion a significant risk of bacteremia to the patients. For example, we feel that there is no need for premedication with penicillin in these cases.

We recommend that teeth be extracted singly when a patient has a history of rheumatic fever or congenital valvular disease. Proper management of such cases still includes the use of penicillin, although its beneficial influence is yet to be definitely established.

SUMMARY

The incidence of transient bacteremia following dental extractions and periodontal scalings was studied in 221 cases. The results obtained justify certain conclusions.

1. The incidence of dental bacteremias was considerably less than that previously reported by other investigators. The over-all incidence of 10 per cent differed significantly from the 40 per cent incidence obtained by combining the results previously reported. One positive blood culture was found among 56 single extractions, and 16 positives among 92 cases of multiple extractions.

2. Bacteremias, when they occurred, were of very short duration. In the majority of cases, bacteria were detected only within the first minute following completion of the operation.

3. The age of the patient, the amount of operative trauma, and the number of teeth involved in the operation exerted definite influence on the incidence of bacteremia. Bacteremias occurred more frequently in patients below the age of 30 and above the age of 60 than they did in patients aged 30 to 59. The incidence was greater when moderate or extensive trauma accompanied the operation than when only slight trauma was involved. Significantly greater numbers of bacteremia occurred following extraction of two or more teeth than following extraction of single teeth.

4. Variations in tooth mobility and vitality and the presence of periodontal disease or apical infection did not appear to influence the incidence of bacteremia.

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AN ADHESIVE PENICILLIN OINTMENT FOR TOPICAL APPLICATION

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THE value of penicillin in the treatment of infections of the gingivae and oral mucosa has been established.[1-5] Topical application gives a higher surface concentration at the site of the infection than does parenteral use, and is, therefore, the treatment of choice for most infections of these tissues.[3, 6, 7]

The vehicle employed is of utmost importance. It must not be incompatible with or detrimental to the antibiotic, nor have any deleterious action upon the hard and soft tissues or restorations. It should not be toxic if swallowed. It must release the penicillin readily. It should be stable under ordinary circumstances, and be free from objectionable taste, odor, or staining. A viscous base which would adhere to the tissues and slowly release penicillin over a period of time would be advantageous. Schautz and Scrivner reported the use of penicillin mixed with a denture adhesive powder[8] for this purpose. The principal ingredient of most denture adhesive powders is Karaya gum. Carboxymethyl cellulose is a substance of similar nature and properties. Being a synthetic product, it is more uniform than Karaya gum, and may be rendered sterile.

Karaya gum and a mixture of carboxymethyl cellulose with petrolatum were applied to the abdomen of rabbits. The field was prepared by shaving, and two applications were made each day for 37 days. Control animals were used. There was no evidence of skin reaction, erythema, glandular involvement, infection, or obstruction of the hairy growth from either of these substances. These bases were fed to rats and mice. The dosage for mice started at 2.5 mg. and was increased to 10 mg. b.i.d. for 8 days. Rats were started at 20 mg. and reached 100 mg. b.i.d. for 13 days. This is equivalent to 6 Gm. per dose for a man of average weight. At the end of the test period, all animals were healthy, showing no reaction, except a slight diarrhea which might be attributed to the volume of liquid material introduced into the stomach.

Crystallin penicillin was incorporated into both bases in proportion of 10,000 units per Gm. Samples were stored at room (23-26° C.), refrigeration (3° C.), and incubation (37° C.) temperature for one month, then tested for potency. The Karaya gum-penicillin mixture stored at 37° C. had completely

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lost its antibiotic power. All other samples retained their antibacterial effect. The Karaya gum base was discarded, therefore, and further studies continued on the carboxymethyl cellulose base.

Samples of this base containing 4,500 and 10,000 units were stored as above. At the end of 7 months, all samples produced definite zones of inhibition on plates streaked with standard stains of *Staph. aureus* (Table I and Fig. 1).

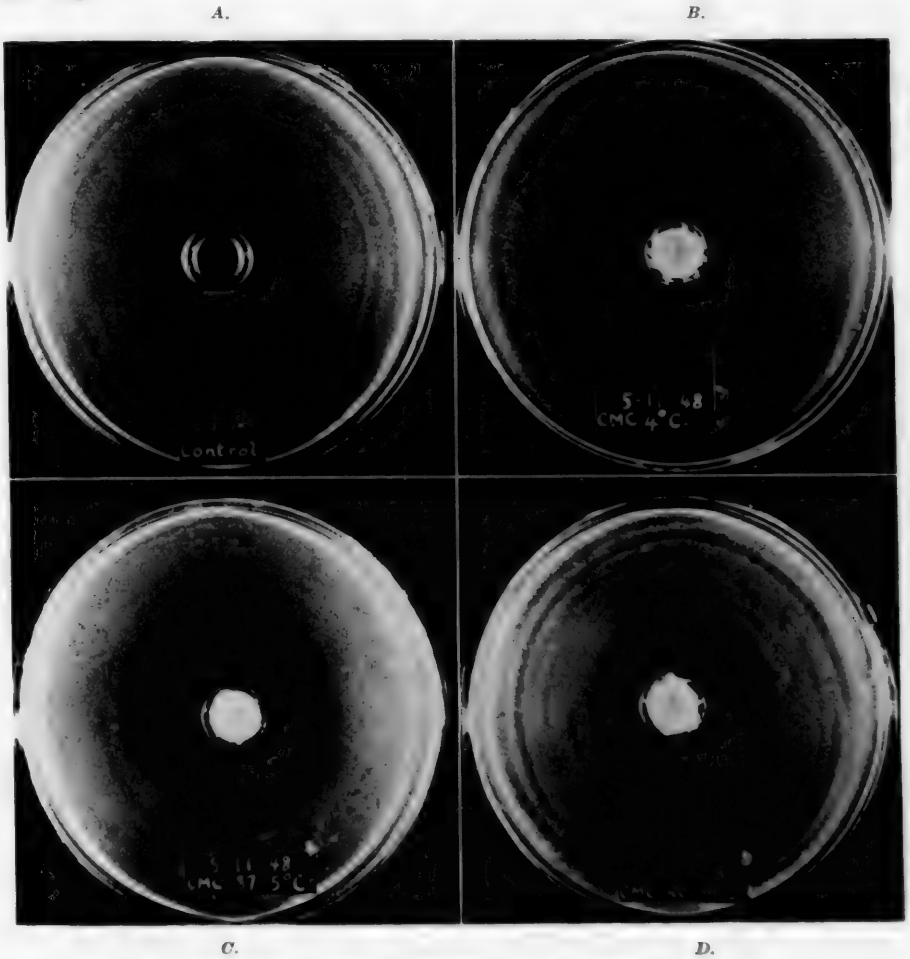


Fig. 1.—Cultures showing zones of inhibition after 7 months.
A. Base only—no zone.
B. Ointment at 4° C.
C. Ointment at 37° C.
D. Ointment at room temperature.

Samples were examined to determine their effect upon the motility and growth of the oral flora. A modification of the Rosenthal motility test[9] was employed. A thin ring of the cream was placed on a warm glass slide leaving

a clear space in the center approximately .5 cm. in diameter. Two loopfuls of fresh saliva from a suitable subject containing the usual bacteria and debris were introduced into the center space. A cover glass was placed over the preparation and examined by dark-field microscope. The time required for

TABLE I
ANTIBIOTICS ACTIVITY OF PENICILLIN OINTMENTS
CMC

SAMPLE 5/11/48	STORAGE CONDITION	6/17	7/16	8/16	9/16	12/16
42 mm.	Incubator	30 mm.	35 mm.	33 mm.	32 mm.	35 mm.
	Room temperature	38 mm.	33 mm.	35 mm.	33 mm.	35 mm.
	Refrigerator	40 mm.	33 mm.	30 mm.	30 mm.	33 mm.
C base M C		neg.	neg.	neg.	neg.	neg.

cessation of motility of spirochetes, vibrios, and motile bacilli was recorded. Because organic and inorganic debris slows the diffusion of medicaments and consequently delays contact with the bacterial cell, specimens were selected containing moderate amounts of debris.

The effect on growth was determined by forming in a Petri dish a ring of cream as above, but approximately 2 mm. high, and filling the center with fresh saliva. This was allowed to stand for 30 minutes; after which 2 loopfuls of the saliva were introduced into nutrient broth, Brewer's broth, and an agar slant. All media were incubated 72 hours and examined. The results (Table II) show that the vehicle exerts no antibacterial effect per se, and that the higher concentration is the more effective.

TABLE II
EFFECT ON MOTILITY AND GROWTH

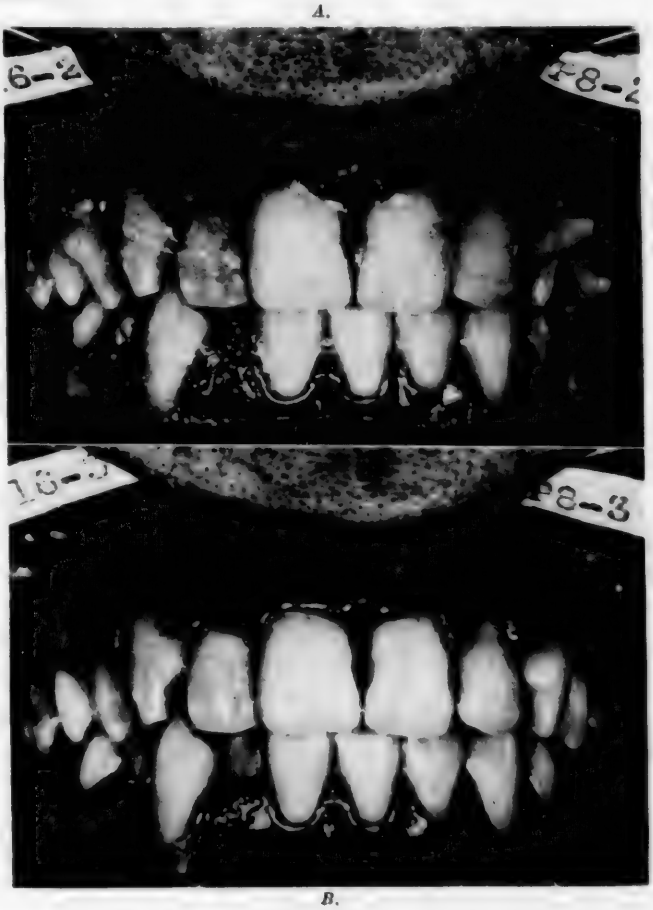
	SPIRO-CHETES	VIBRIOS	BACILLI	NUTRIENT BROTH	BREWER'S BROTH	AGAR SLANT
CMC without penicillin	15	15	15	+	+	+
4,500 units per Gm.	2	2	7	+	+	+
10,000 units per Gm.	1	1	5	0	0	8 colonies

Time for loss of motility is recorded in minutes; 15 = still motile at 15 minutes, + = growth, and 0 = no growth.

Both concentrations were tested clinically using 3 Gm. of the 4,500 unit ointment or 1.5 Gm. of the 10,000-unit mixture. Debris was removed by a saline spray under pressure. Material was taken from the gingival sulcus at the site of a lesion, and examined by stained smears and by dark-field examination. The penicillin ointment was then applied to all lesions and packed into all interdental spaces. At intervals of 15, 30, and 60 minutes, the ointment was thoroughly removed from the same and one additional site, and material again removed for study. The ointment was then replaced. The ointment remained in considerable quantities for the entire period. When 1.5 Gm. of material were used, some remained for as long as 3 hours. When 3

Gm. were used, some remained for 5 hours; but patients complained of the excessive bulk.

Twenty-five cases were treated: 22 acute Vincent's infection, 2 pericoronitis, and 1 acute streptococcic gingivitis. All acute infections were successfully controlled. In most instances, pain was relieved within 2 hours. Fusospirochetal organisms disappeared and other bacterial forms were reduced in number.



A.

B.

FIG. 2.—Acute Vincent's infection.

A. Patient one hour after application of penicillin ointment. The material is still in position and in considerable quantity, a small portion has been lifted away from between the first incisors to expose necrotic area. Note amount of inflammation.

B. Forty-eight hours later. Patient is comfortable, inflammation is reduced.

CASE REPORTS

Since the bacteria were too numerous to count accurately, they are designated: ++++ = countless numbers in every field; + = a few scattered bacterial cells in occasional fields; ++ and +++ = numbers in proportion. Speed of motility is similarly represented. These cases are typical of the group.

CASE 1. (FIG. 2) ACUTE VINCENT'S INFECTION. CMC 10,000 UNITS PER GM.

Motility

MINUTES	SPIROCHETES	VIBRIOS	MOTILE BACILLI	COCCI
0	++++	++++	++++	----
15	++	+	++	----
30	0	0	+	----
60	0	0	0	----
24 hrs.	0	0	++++	----

Numbers

MINUTES	SPIROCHETES	VIBRIOS	MOTILE BACILLI	COCCI
0	++++	++++	++++	+
15	++	+	++	++
30	0	0	+	+++
60	0	0	0	++
24 hrs.	0	0	++++	+++

Relief of symptoms and improvement in color in gingivae.

CASE 2. ACUTE PERICORONITIS

Motility

MINUTES	SPIROCHETES	VIBRIOS	MOTILE BACILLI	COCCI
0	++++	++++	++++	----
15	+	0	+++	----
30	+++	0	++	----
60	0	0	0	----
24 hrs.	++++	0	++++	----

Numbers

MINUTES	SPIROCHETES	VIBRIOS	MOTILE BACILLI	COCCI
0	++++	++++	++++	++
15	+	0	++	+++
30	+	0	+++	+++
60	0	0	0	++
24 hrs.	+	0	+++	+++

Complete relief from pain.

SUMMARY

Carboxymethyl cellulose with petrolatum is a suitable vehicle for the topical application of penicillin in the treatment of Vincent's infection and similar infections of the oral mucosa. Preferably, it should contain 10,000 units of crystallin penicillin per gram.

The base alone has no antibacterial properties, but is not toxic and is compatible with crystallin penicillin. It can be safely stored at room temperature for at least 7 months.

The ointment will remain in situ for several hours, i.e., longer than necessary to exert its antibacterial action.

Its use does not obviate the necessity of proper prophylactic care in these cases.

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THE ORGANIC ELEMENTS OF THE ENAMEL

II. The Organic Framework of the Internal Part of the Enamel, With Special Regard to the Organic Basis for the So-Called Tufts and Schreger's Bands

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IT is generally agreed that the morphology of most tissues of the human body has been studied so extensively by conventional histological technics that significant progress could hardly have been envisaged were it not for the development of several new scientific tools. Radiochemical, histochemical, and submicroscopical observations, to mention a few, have recently begun to breathe new life into the morphological pattern of an ever-increasing number of biological structures.

Because intelligent application of these newer tools requires exact morphological orientation, it is, more than ever before, necessary to explore fully the morphology of those structures about which there is still something to be learned by the power of the ordinary microscope, by binocular observations, and microdissection. The enamel of the teeth is believed to fall in this category, due, in part, to technical obstacles unique to this structure.

This applies in particular to the organic elements of the enamel. The development of our knowledge in this field,[1] so essential to the progress of preventive dentistry, has, moreover, been hampered by the fact that some histologists have questioned the very existence of an organic framework in the enamel of mature human teeth.

An analysis of the various technical difficulties encountered in this problem focused attention upon some of the principal factors involved in the successful isolation of the organic framework of mature enamel.[2] Preliminary success in preparation of gross specimens, smears, and paraffin sections of demineralized enamel made it possible to give a brief account of the three-dimensional appearance of the organic framework.[3] The following observations are concerned with the gross architecture and histomorphology of the organic elements which are most characteristic of the internal portion of the enamel and, more particularly, with the organic basis for the so-called tufts and Schreger's bands.

PROCEDURE

The problems of immobilization and decalcification which, from the first study in this series,[2] appeared to be crucial factors in the successful isolation of the organic elements of the enamel were solved by a comparatively simple technic in the present study which, at this time, did not aim at specific histological or histochemical detail.

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Demineralization was achieved primarily by the use of low-temperature decalcification with 5 per cent trichloro-acetic acid, using adult human teeth which had been preserved in 10 per cent formalin for a period of a week to a few months preceding decalcification. In order to minimize destructive CO_2 formation, the decalcification process was carried out inside an icebox at 1° Centigrade. Each specimen was immobilized in an individual celloidin bag.*

When the teeth had been exposed to the acid for about an hour, the surface cuticle could be seen as a thin transparent membrane which appeared to become slightly elevated from the enamel surface. In the course of from two days to two weeks, depending upon the type and size of the specimen, the enamel had been demineralized to a point where it looked like a transparent cap, so delicate that it could easily be torn apart if carelessly handled. When the enamel was the main object for further preparation, which was the case in this particular series of observations, the celloidin bag containing the tooth and decalcification fluid was at this point carefully lifted out of the decalcification vessel and into a jar containing cold water. Still under low temperature, the specimen was washed free from acid by carefully lifting the celloidin bag from one cold water bath to another. During this process it proved important to avoid shaking, and to ascertain that the specimen was continuously surrounded by fluid, in order to prevent tearing or collapse of the delicate organic framework of the enamel.

Under binocular microscope, and with the specimen submerged in water and strongly illuminated, the surface cuticle and the homogeneous-looking, cloudlike external organic framework could now be dissected away from the internal part of the organic framework of the enamel.[2] The latter part was by comparison fairly dense and rigid, and could be separated and elevated from the dentin by means of a thin needle, such as a No. 14 smooth root canal broach. This separation was simplified by the finding that the internal framework rested on a fairly solid basal membrane, which often could be removed in one piece (compare Figs. 4 and 11).

Kept under fluid, such specimens were studied grossly under binocular microscope, then dissected into separate specimens for special microscopical study as smears and paraffin sections.

In addition to these teeth which were decalcified individually, it was, for certain purposes, desirable to decalcify simultaneously as many as 50 to 100 teeth under identical conditions. This gave an opportunity to determine which specimens could be considered typical for the architecture of the organic framework. Furthermore, it was thus possible to dissect and collect in separate vials quite large and noncontaminated quantities of special fractions of the organic elements, such as the surface cuticle, lamellae, external and, for the present purposes, the internal portion of the organic framework.

*The celloidin bags were made by wetting the inside wall of one-half inch diameter glass vials with a thin coating of 2 per cent celloidin which, after a minute's drying, could be removed from the glass wall by immersing the vial in water.

RESULTS

Following the removal of the surface cuticle, a layer has regularly been found in the internal part of the enamel which contains a greater abundance of organic matter than the external part of the enamel. An illustration from one of the first and crudely recorded observations in this series of studies is shown in Fig. 1. In this specimen most of the surface cuticle, as well as the external part of the organic matter of the enamel, has been lost. This is best seen from the profile of the molar cusps at the top of the picture. In the center, where most of the dentin was drilled away, the tooth is sufficiently transparent in transmitted light to show several rows of demineralized organic remains. Under somewhat higher magnification (Fig. 2), the stripes are seen to be fairly regularly spaced. When smears are taken from this region, one can see some structures which give the impression of being patent and branching in several directions (Fig. 3).

The following extract from one of my early decalcification experiments records the gross appearance of the internal organic structures of the enamel:

> A mandibular premolar was successfully split so as to separate the buccal shell of enamel from the dentin. Decalcification of the specimen was observed under the binocular microscope. On the enamel surface facing the dentin a white cloudy material came into view as the salts were removed. After a while it became possible to distinguish a white network, the meshes having a width of less than one-half of a millimeter. Of the 4 walls of these large meshes the two running in the longitudinal direction of the tooth were of a heavier nature and resolved themselves into white bundles of a wavy character. A smear made from this region suggested that these organic remains included elements of a tubular and branching character and of a diameter exceeding that of an average enamel prism.

It would not seem surprising if the delicate structures described above were regularly lost during washing, dehydration, or embedding of decalcified teeth, if not completely disrupted by the decalcification procedure itself. Following these early observations, greater precautions were, therefore, taken during decalcification of the specimens. Instead of taking small individual smears, it became possible through more experience to dissect and isolate whole areas of this internal part of the organic framework of the enamel. This portion could then be examined separately and in toto. Figs. 4 and 5 show the full length of the buccal portion of such a specimen taken from a first permanent molar. Photographed while the specimen was kept in water, Fig. 4 shows, in reflected light, the wavy (corrugated) character of the structures. In Fig. 5 the same tissue is shown in transmitted light, in which case it is noted that the regularly spaced wavy structures transmit much less light than the areas in between.

When a small segment of the internal organic framework is dissected free and observed at a somewhat higher magnification, it is easier to follow the wavy course of the organic remains, both in reflected light (Fig. 6) and in transmitted light (Fig. 7). It is obvious, furthermore, that these regularly spaced rows of corrugated structures run in the longitudinal direction of the tooth. When turned at right angles, and balanced on the edge* the same speci-

*This was achieved by supporting the specimen between the thin edges of two glass slides, resting on the bottom of a dish of water, which contained the specimen to be photographed.

Fig. 1.

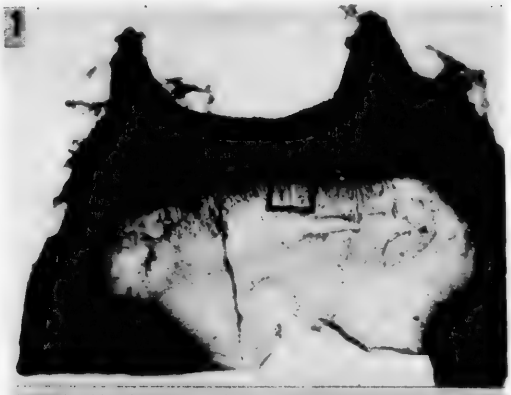

Fig. 2.

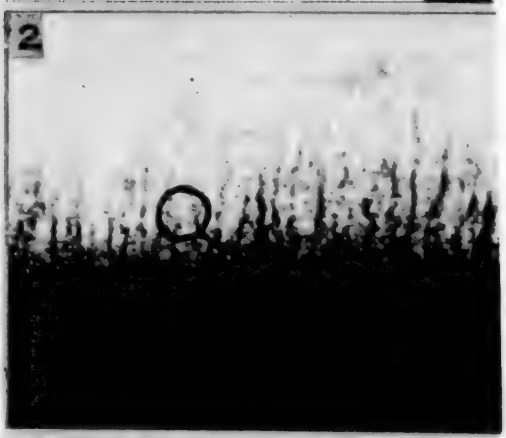

Fig. 3.

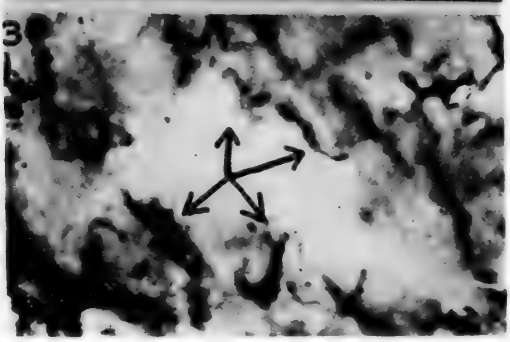

Fig. 1.—Adult human molar, decalcified after removal of most of dentin. Remnants of internal enamel framework show longitudinal rows of organic matter. Unstained gross specimen, immersed in water, and photographed in transmitted light. ×5.

Fig. 2.—Region indicated by the square in Fig. 1. ×10.

Fig. 3.—Smear from region indicated by circle in Fig. 2 suggesting the presence of patent, branching structures in internal organic portion of enamel. ×1000.

Fig. 4

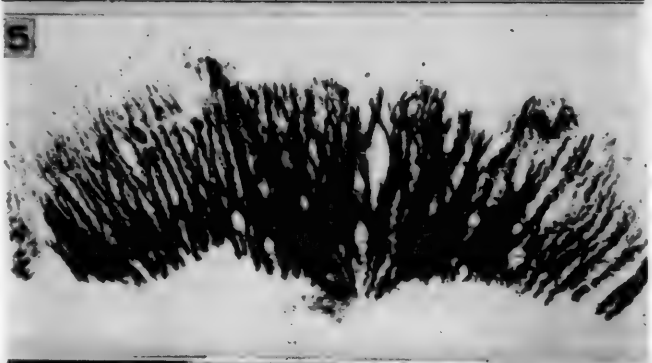

Fig. 5.

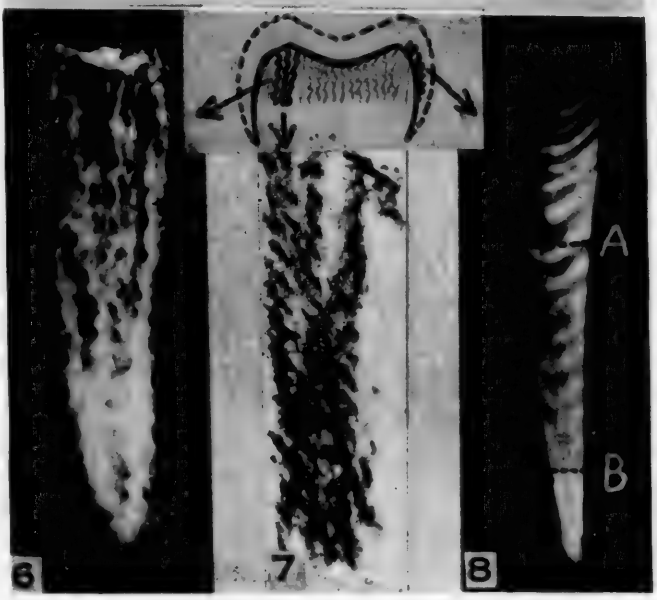

Fig. 6. Fig. 7. Fig. 8.

Figs. 4 to 8.—Whole segments of unstained internal organic framework of enamel removed by binocular dissection from decalcified adult human molars. Isolated specimens were submerged in water, and photographed in light reflected from side (Figs. 4, 6, 8) and transmitted from below (Figs. 5, 7), respectively.

Fig. 4.—Internal organic portion of buccal shell of enamel. Organic matter is divided into regularly spaced longitudinal wavy rows, giving a corrugated pattern. ×15.

Fig. 5.—Same as above, seen in transmitted light, suggesting much denser accumulation of organic matter in alternating longitudinal layers. ×15.

Figs. 6 and 7.—Higher magnification of small segment of above, seen in reflected and transmitted light, respectively, and confirming corrugated and dense character of longitudinal plates of internal organic framework of the enamel. ×30.

Fig. 8.—Segment shown in Fig. 6 turned 90 degrees, that is, seen from side in reflected light. This picture shows light and dark bands strikingly similar to Schreger's bands which are familiar in undecalcified longitudinal ground sections of enamel. ×30.

men can be viewed from the side (Fig. 8). This corresponds to a view of a longitudinal plane and gives the pattern of light and dark bands, significantly similar to the pattern of Schreger's bands, which indicates that the appearance of these bands, previously familiar from ground sections, is brought about by the pattern of the organic framework of the enamel.

In order to identify the microscopical pattern of the longitudinal wavy bundles of organic matter, it is necessary to make sections in different planes. A section through the plane marked *A* in Fig. 8, where the Schreger's bands are most predominant, is seen in Fig. 9. This section shows several rows of regularly spaced denser bundles of prism sheaths running a wavy course. Between these are rows of less densely packed and straighter running bundles of prism sheaths, resting on a partly retained scalloped basal membrane. In the center of each of the denser bundles are seen somewhat darker lines which, in higher magnification (Fig. 10), appear to be patent structures, possibly corresponding to those noted in the smear described before (Fig. 3). This central portion of the corrugated longitudinal segments of the internal enamel is, furthermore, believed to be the main component of the brushlike structures (tufts), which break the light when transversely cut ground sections are observed in a single plane.

Fig. 11 shows a section through the plane marked *B* in Figs. 4 and 8, in which the wavy nature of the bundles and the Schreger's bands, respectively, are less marked. This region exhibits fewer and less marked tufts, and more evenly distributed prism sheaths resting on a straight basal membrane *(membrana limitans)*.

Where the enamel is fairly thick, as is generally true as one approaches the occlusal ridge of the molars, a more marked corrugation of the internal organic framework of the enamel was generally found. A typical example is shown in Fig. 12, from which specimen the external homogenous portion has been removed by dissection. Similarly, it was found that the dentin, upon which such corrugated enamel was resting, often showed a marked scalloping, an extreme example of which is shown in Fig. 13.

As a rule, the prisms become wider and take a fairly straight course before reaching the wider external curvature of the enamel surface. Thus, it should be emphasized that the picture seen in Fig. 12, which is strikingly suggestive of an enamel surface exhibiting many coarse imbrication lines, actually represents a region far below the enamel surface, namely, the point of transition from the internal and corrugated portion to the external and homogenous portion of the enamel framework. In this instance the corrugation, the tufts, the Schreger's bands, as well as the pattern suggestive of imbrication lines, were, as a matter of fact, lost by the time the prisms reached the external curvature of the enamel. Indeed, even on top of the extremely uneven dentin surface, seen in profile in Fig. 13, could be found a shell of enamel which, at its thickest points had "straightened out," so to speak, to form a homogenous external layer and, consequently, a relatively smooth surface contour of the enamel.

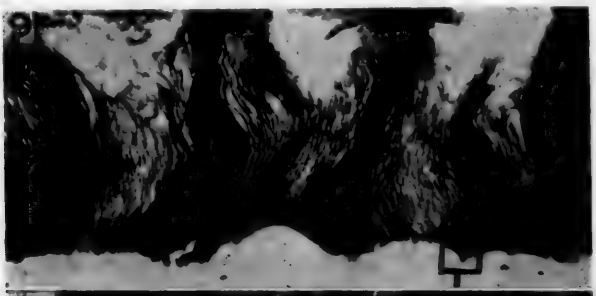

Fig. 9.

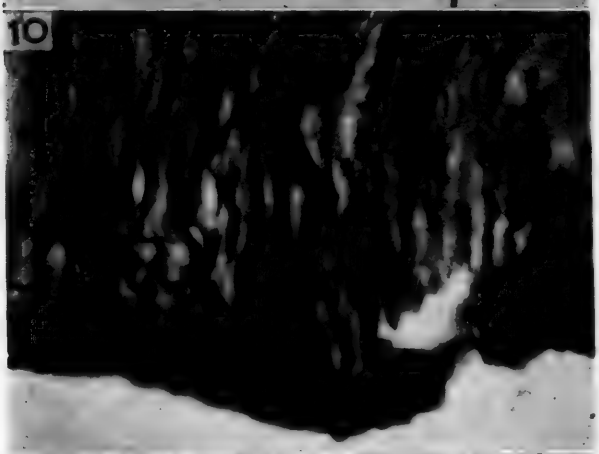

Fig. 10.

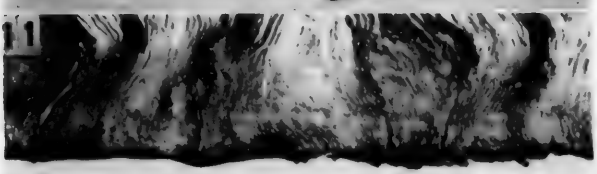

Fig. 11.

Fig. 12.

Fig. 13.

Fig. 9.—Paraffin section through the region marked *A* in Fig. 8. Basal membrane has been successfully separated from an elevated point in dentin surface from which this specimen was separated. In central portion of dense prism bundles are seen one or more light-absorbing lines. H. & E. stain. ×200.

Fig. 10.—Higher magnification of "lines" mentioned in Fig. 9, suggesting patency similar to structures seen in the smear from corresponding region described in Fig. 3. These central organic structures, which tend to absorb or break transmitted light, are believed to be mainly responsible for the appearance of the so-called tufts in ground sections. ×1650.

Fig. 11.—Paraffin section through region marked *B* in Fig. 8. Note straight course and complete preservation of basal membrane, absence of dense, wavy prism bundles, and scarcity of darker (patent) structures. This homogenous framework lacks organic pattern responsible for tufts and Schreger's bands. H. & E. stain. ×200.

Fig. 12.—Gross specimen of internal organic framework of enamel covering buccal surface region of an adult human molar cusp. (External homogenous framework has been removed by microdissection.) Unstained specimen, submerged in water and photographed with light reflected from surface. ×10.

Fig. 13.—Profile of buccal plate of dentin of a Pecos Indian's tooth, showing an extremely scalloped dentin surface, formation of which precedes and, consequently, is believed to influence formative pattern of organic framework of enamel.

COMMENTS

The fact that the full length of the straight running basal membrane, referred to in Fig. 11, had been successfully separated from the dentin following the decalcification, would seem to indicate that the said membrane is more intimately connected with the enamel matrix than with the dentin matrix. The straight course of the membrane is, moreover, believed to explain the fact that the enamel prisms in such a case proceed on a fairly straight course from a relatively smooth dentin surface, finally to terminate in a similarly straight and smooth enamel surface.

Where, on the other hand, an uneven, scalloped dentin surface exists, the similarly uneven basal membrane of the enamel matrix is correspondingly harder to separate from the dentin, as indicated by comparing Figs. 9 and 11. It is also evident from Fig. 9 that the deeper points, or indentations, in the dentin surface serve as the starting point for the regularly spaced, dense, and wavy bundles of prism sheaths.

Where the shell of enamel is fairly thin, such as in the cervical region of certain teeth, the uneven dentin surface and the wavy course of the prism bundles, that is, the corrugation of the enamel, may reflect itself all the way to the enamel surface. It is possible that this condition has a bearing upon the prevalence of the coarse imbrication lines generally found on the surface of the thin cervical enamel, which is characteristic of the permanent teeth of man.

The above findings, and the fact that the enamel, embryologically, is known to be preceded by a rigid rim of dentin, would seem to suggest that the external architecture of the dentin may permanently reflect itself in the morphology of the enamel, occasionally even in its surface texture but, particularly, in its internal organic framework, its degree of corrugation and, consequently, in the optical characteristics known as tufts and Schreger's bands.

SUMMARY

The internal portion of the organic framework of adult human teeth has been studied, after low-temperature demineralization, by means of binocular observations, microdissection, smears, and paraffin sections.

In contrast to the more homogenous external portion, with its wide and straight-running prism sheaths, the internal organic framework is characterized by densely packed prism sheaths, which are arranged in longitudinal corrugated segments. Viewed in a transverse plane the central part of these denser segments gives rise to the appearance of the so-called tufts, and in a longitudinal plane the corrugated pattern will reflect itself in the grossly visible Schreger's bands.

It is suggested that the morphology of the external dentin surface, which embryologically precede the formation of enamel, may have a bearing upon this pattern of the organic framework of the internal enamel, and may even be

reflected in the surface texture of the enamel in cases where a scalloped dentin surface is covered with only a thin shell of enamel.

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THE ORGANIC ELEMENTS OF THE ENAMEL

III. The Pattern of the Organic Framework in the Region of the Neonatal and Other Incremental Lines of the Enamel

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ON THE basis of studies of ground sections of the enamel, earlier workers seem to have concurred in the belief that the incremental lines or striae of Retzius (1837) are due to a faulty, poor, or low degree of calcification (Lehner and Plenk, 1936). Conclusions differ as to whether the presumably deficient calcification involves the enamel rods, as well as the inter-rod substance, or is limited to the rods, their periphery, cross striations, or inter-rod substance.

Noyes, Schour, and Noyes (1948) stated that "A striae of Retzius represents a poorly calcified portion of the enamel, because it is made up of the relatively less calcified components of an enamel rod." If this were correct one might perhaps expect that a complete decalcification of the whole enamel should tend to equalize or eliminate this difference which is believed to exist between regions of a high ("normal") and a low ("poor") degree of calcification. This is precisely what appears to have been experienced by Frisbie, Nuckolls, and Saunders (1944) who, after having studied decalcified sections of enamel, reported that "incremental and striation lines have never been observed when the calcium salts are totally removed." The latter conclusion, if confirmed, would seem to relegate to an artifact an illustration by Bodecker (1911) which in the past appears to be the only significant exception to the above generalization. No reference to Retzius lines was made in Bodecker's original publication, but when recently reproduced in the textbook of Orban (1944) the said illustration was suggested to show that a line, simulating the striae of Retzius, had been recovered in the organic framework of enamel after decalcification in acid celloidin. One may perhaps question the efficacy of such a viscous and comparatively low-dissociating mixture in completely removing calcium salts, in spite of the long period of time (1 to 6 months) expended for the softening of a small piece of enamel by that method. Nevertheless, there are in the field of enamel histology many sources of artifacts which may be to blame for discrepancies of this nature.

Taking advantage of somewhat different technics, the principles of which have been discussed in detail elsewhere (Sognnaes, 1948), observations have been made which are believed to indicate that the striae of Retzius, as well as their accentuated forms, such as the neonatal lines, actually owe their appearances to the pattern of the organic framework of the enamel and more specifically, if not entirely, to the morphology of the organic sheaths surrounding the enamel prisms.

Received for publication, February 24, 1949.

PROCEDURE

Decalcified paraffin sections were prepared from caries-free deciduous molars removed from clinic patients just prior to the time of normal exfoliation. Having thus obtained teeth which had served their complete functional period without becoming decayed, the present material is believed to represent as mature and intact enamel as can be obtained for the study of neonatal and other incremental lines.

Following a few days' submersion in 10 per cent formalin, the teeth were prepared for paraffin sections by means of a simple decalcification procedure which may be referred to as a low-temperature "fix-as-you-go" technic.[12] The method consisted of a simultaneous fixation and decalcification in a solution of trichloroacetic acid (5 Gm.) and potassium bichromate (2.5 Gm.) dissolved in distilled water (100 c.c.) to which was added formaldehyde (10 c.c.). Each tooth was inserted separately in small formalin-filled celloidin bags (made by coating the wall of an 0.5-inch glass tube with a 2 per cent solution of celloidin), placed in a beaker containing the above solution, and kept in an icebox at 1° Centigrade. The celloidin bags served three purposes, the need for all of which was evident from a previous report[12] in this series: first, to slow down the initial penetration of the acid; second, to immobilize the specimen; and third, to keep it continuously surrounded by fluid when transferring it from one solution to the other. During a week's submersion in the above-mentioned solution, the organic elements of the enamel were slowly exposed and thus simultaneously decalcified, fixed, and hardened. The specimens, still contained in the separate celloidin bags, were then transferred to a 5 per cent water solution of trichloroacetic acid for an additional length of time, depending upon the amount of dentin but, in the present cases, not exceeding two weeks.

Next the specimens, still in the celloidin bags, were submerged in cold water for several washings, and carried carefully through increasing concentrations of ethyl alcohol to butyl alcohol. From this solution, which is miscible with paraffin, the dehydrated specimens could be lifted into paraffin by means of a small spoon which had been made for that specific purpose. In this manner it was possible to keep the delicate enamel framework continuously in a fluid medium in order to prevent its collapse, tearing, or loss. Having thus carefully transferred the specimens to paraffin, sections were cut in routine fashion, deparaffinized, and treated with hematoxylin-eosin, Azan, and other standard stains.

The fact that the paraffin sections contain dentin, as well as enamel, means that the decalcification was carried beyond the stage necessary for decalcification of the enamel alone.[5]

Cautioned by the past history of our histological knowledge of enamel, it should not be overlooked that some extremely delicate or as yet unrecognized components of the organic framework of the enamel may well have been lost in the specimens to be presented because of the prolonged decalcification or the still existing deficiencies of our tools. It is unlikely, on the other hand, that

those organic elements which have been recovered could have been falsely retained due to an incomplete removal of calcium salts[6]; this is the more important point to make clear before presenting the results.

RESULTS

Due to the unexpected photographic similarity between the general pattern of the decalcified sections and that of previously familiar ground sections, it was comparatively easy to identify the main morphological characteristics in the paraffin sections of the deciduous molars shown in Figs. 1 to 4. In the center of the enamel in Fig. 1 is a fairly heavy line *(B.L.)* and a number of fainter lines *(R)*, all of which are running nearly parallel to the underlying dentin *(D)*. The former heavy line corresponds in appearance and position to the neonatal line or birth line, which, as far as is known, has not formerly been demonstrated in the organic framework of such decalcified preparations. Like the fainter lines of Retzius *(R)*, it is evident that these incremental lines owe their appearance to the morphology of the main organic elements of the enamel (the prism sheaths) and that the lines follow closely the pattern of contemporarily developed enamel.

This is in sharp contrast to the wide and irregular band of organic matter *(L)*, which is fairly typical of the unpredictable enamel lamellae, characterized by their independence of the course of individual prisms on the one hand, and the course of the incremental lines on the other. In this respect they significantly depart from all of the developmental characteristics of the enamel including the so-called tufts[13] with which they sometimes may be confused when viewed in ground sections.[7]

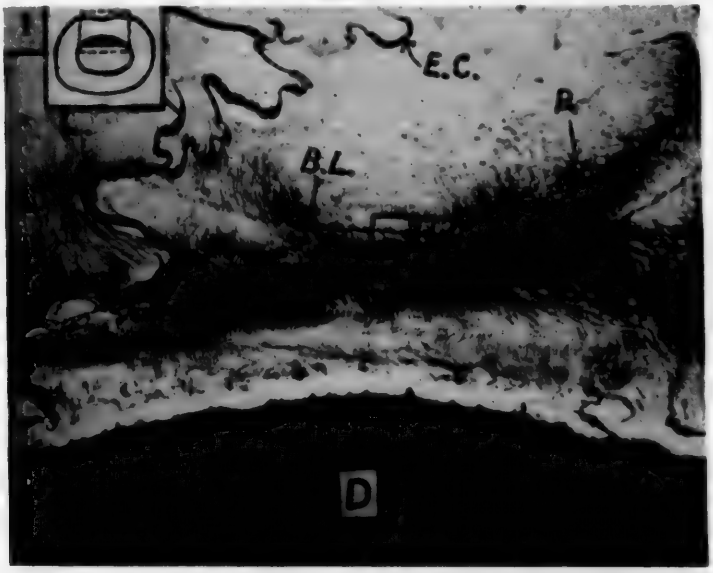
The more minute details of the incremental lines may best be observed in one of their accentuated forms, the neonatal line, which is shown at higher magnification in Fig. 2. This segment of the neonatal line shows that the line is due to somewhat heavier or wider zones in the prism sheaths than exists in the surrounding framework. A clearer picture of this is seen in the longitudinal view in Figs. 3 and 4. These paraffin sections show the cervical portion of a deciduous molar, where the resorption prior to exfoliation has gone beyond the dentin and has begun to produce a resorption line in the internal enamel (*E.R.* in Fig. 3). Only the gingival part of the enamel cuticle is seen on the external surface of the tooth *(E.C.)*. The heavier line *(B.L.)* in the central part of the enamel is in the typical position of a neonatal line, which could easily be confirmed by comparison with ground sections from similar teeth.

In higher magnification (Fig. 4) a segment of the neonatal line shows that it consists of characteristic thickenings of each prism sheath along a line corresponding to contemporary enamel formation. These small thickened areas form a demarcation line between what is considered to be postnatal enamel in the external part and prenatal enamel in the internal part *(P.N.E.)*.

In contrast to the appearance of the incremental lines in thick ground sections is the clear definition of the lines in the thin paraffin sections. Even the accentuated neonatal lines, which at low power and in thick sections appear as one unbroken line, are seen to be built up of separated thickened portions

of the prism sheaths. In higher magnifications it is noted furthermore that not only is the line built up of separate units of the prism sheaths, but several sheaths or certain portions of the prism envelopes are relatively unaffected; thus,

Fig. 1.



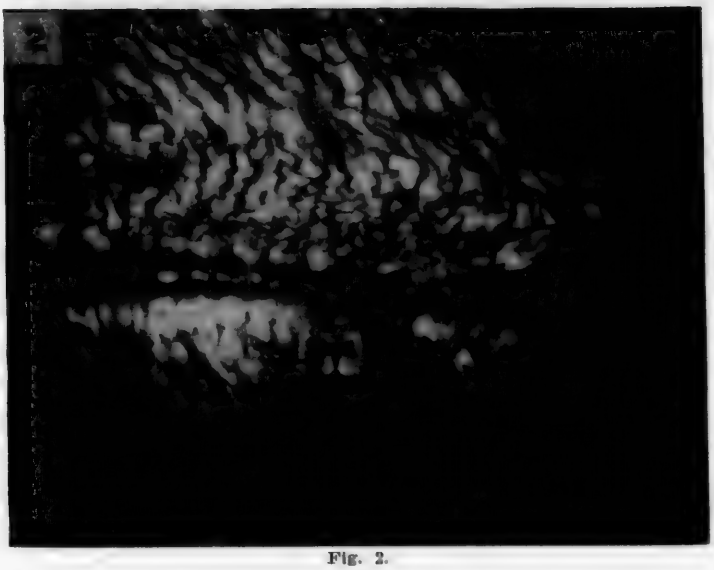


Fig. 2.

Figs. 1 to 4.—Paraffin sections of decalcified deciduous molars extracted after completion of their functional period.

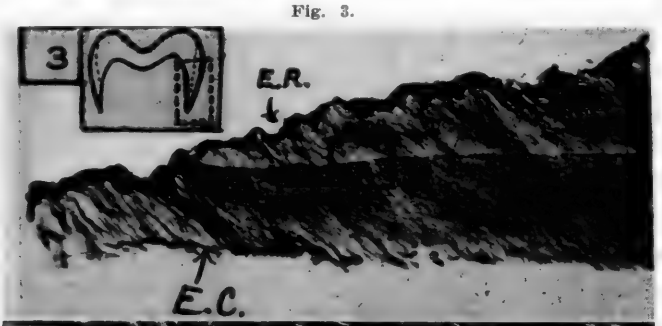
Fig. 1.—Birth line (*B.L.*); Retzius lines (*R*); lamella (*L*); enamel cuticle (*E.C.*); and dentin (*D*). (Azan stain, ×20.)

Fig. 2.—Higher magnification of a segment from Fig. 1, indicating heavy zones of prism sheaths in region of neonatal line. (Azan stain, ×200.)

in Fig. 4, some sheaths only show a slight thickening compared to others in the same line, and in Fig. 2 it is noted that only part of the obliquely cut prism envelopes are involved. Along the straight walls of the envelopes, the sheaths are quite thin, whereas the corners show the most marked thickening. If a section is cut in the long direction of the prisms, it becomes obvious that different regions of the prism envelopes may be seen as more or less heavy zones.

If a number of prisms are within the section, which is usually true in ground sections, there are many combinations in which thickened portions of certain prism sheaths may appear, as if they were intraprismatic rather than interprismatic.

Fig. 3.



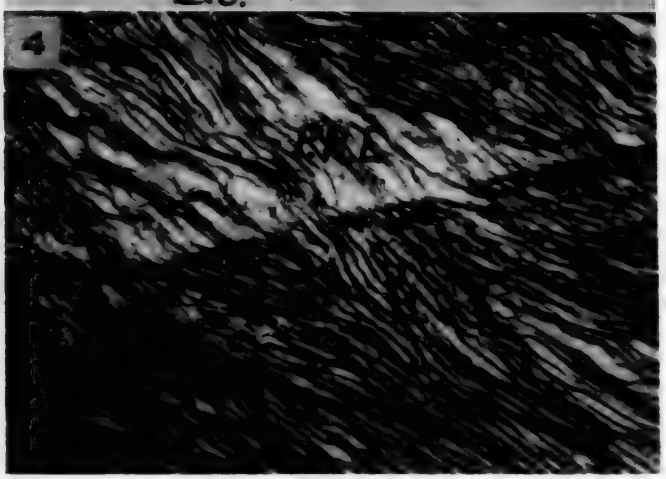


Fig. 4.

Fig. 3.—Mesial part of enamel showing enamel resorptions (*E.R.*); part of enamel cuticle (*E.C.*); and birth line (*B.L.*). (Hematoxylin-eosin stain, ×20.)

Fig. 4.—Segment from Fig. 3, illustrating heavy zones of prism sheaths, i.e., the neonatal line, which segregates prism sheaths of postnatal enamel from the more delicate prism sheaths of prenatal enamel (*P.N.E.*). (Hematoxylin-eosin stain, ×200.)

DISCUSSION AND CONCLUSIONS

Gustafson (1944) has recently suggested that the striae of Retzius, instead of being dependent upon organic substance in the enamel, are simply due to regular bends and changes in the direction of the prisms which, he believes, are caused by traction during the growth of the tooth.

Generally speaking, it would seem difficult to comprehend how the incremental lines, which often are quite broad, can be caused simply by a change in the direction of the prisms. If that were the case, one would either expect a number of small bendings of the prisms which together would tend to break the transmitted light, or one very marked or sharp bend which, if actually present, would lead the prism astray into a quite different path far away from its original direction. Indeed, examination of a great number of teeth seems to indicate that the prisms more often than not go fairly straight through such wide striations and rarely show many small nor single sharp bends. The same is true in the regions of the neonatal lines, and both these and other accentuated striae seem to be quite satisfactorily explained by an unusually long zone or several closely spaced zones of the wide prism sheaths shown by the accompanying decalcified sections. It may also be noted that Gustafson's hardness tests, which gave similar results in and around areas of striated enamel, were too crude to test separately the hardness of the isolated regions of the prism sheaths per se which, as shown above, are more intimately related to the striae of Retzius.

However, if it can be demonstrated that the prism sheaths in the region of the Retzius lines actually are as evenly hard and calcified as the prisms which they surround, then obviously the whole prism sheaths, including their incremental zones, must eventually become completely petrified by highly calcified matter. This is, of course, entirely possible in fully mature teeth, and yet this situation would not necessarily contradict the following conclusion: first, that the organic framework has a pattern of its own in the regions of the incremental lines, which (like the organic framework of the highly mineralized sponges and corals) can be successfully recovered if carefully demineralized; and second, that the recovered organic elements satisfactorily explain the morphological appearance of the incremental lines of the enamel, whether they be accentuated or not, and whether they be viewed in ground sections or decalcified sections.

It is believed that the "bead-like" widenings between the prisms suggested by Abbott (1887), the "pearl-string" appearance of the prism sheaths mentioned by Andresen (1902), the "clefts" noted by von Ebner (1905), and, finally, the dotted line along the prism sheaths referred to in the previously mentioned illustration by Bodecker (1911) all have been conveyed by the same thickenings of the prism sheaths which are so characteristic in the illustrations of the decalcified paraffin sections of the enamel shown in the present study.

SUMMARY

The morphology of the incremental lines of the enamel has been studied in paraffin sections prepared by a low-temperature decalcification procedure.

The optical appearance of the incremental lines of the enamel, familiar from earlier studies of ground sections, is mainly caused by the morphology of the organic envelopes surrounding the prisms. In decalcified sections of deciduous teeth the striae of Retzius and the neonatal lines were found to be composed of a number of heavy zones in the prism sheaths along regions of contemporary enamel formation.

This investigation was supported in part by a research grant from the Division of Research Grants and Fellowships of the National Institutes of Health, U. S. Public Health Service.

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DENTINO-ENAMEL CUTICLE PRESENT IN ADULT HUMAN TEETH

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THE presence of the dentino-enamel cuticle prior to dentinogenesis and amelogenesis is generally recognized by several investigators. Orban, Sicher and Weinmann[1] expressed the opinion that this cuticle is the result of the fusion of the basement membrane of the dental papilla and the fused basal ectoplasmic zones of the ameloblasts. Whether this cuticle persists in adult teeth after having undergone the maturation process is a question which, apparently, is not yet definitely answered. From the available literature at our disposal, the writer was unable to find any report showing the presence of the dentino-enamel cuticle in adult teeth. Standard textbooks in oral histology seem to be silent also about the existence of this cuticle in their description of the organic matter found at the dentino-enamel junction. It is, therefore, on this account that this report is made. Its findings might be of some significance to our future studies in dental histology and pathology.

MATERIALS AND METHOD

Decalcified sections of two teeth were used in this report: one is a partially erupted lower third molar with a lesion of caries and the other, an upper molar without evidence of caries in the enamel. These teeth were extracted from different individuals who were over 30 years of age.

The method employed in the preparation of the histologic sections is that recommended by Willman,[2] with some modification. The lower third molar was decalcified in 5 per cent nitric for twenty days instead of seven to nine days as recommended. The acid solution was changed once after ten days in order to insure, more or less, complete decalcification of the specimen. The upper second molar was only decalcified for nine days. In embedding, only one solution of the celloidin (12 per cent) was used. The thickness of the sections is about 16 microns.

FINDINGS

Fig. 1 is of a longitudinal section of the lower third molar which was decalcified for twenty days. A portion of the enamel matrix in the occlusal region is recovered and undergoing a carious process similar to that described by Gottlieb[3] and by Frisbie, Nuckolls, and Saunders.[4] As shown, a membrane-like structure is present partly detached from the dentin surface and attached to the enamel matrix. When completely free from debris of the necrotic matrix, this membrane appears just as a mere line (Fig. 2). It is possible that this membrane may be the "coagulated tissue fluid" mentioned by

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Gottlieb[5] in his discussion on undermining caries. As shown in the illustration, there is unmistakably an undermining caries above the membrane-like structure as indicated by the disappearance of some of the necrotic enamel matrix. Fig. 3, however, is a section of the upper molar without any evidence of a carious lesion showing an identical membrane-like structure which was completely detached from the dentin surface during histologic preparation. It

Fig. 1.—Decalcified section of a lower third molar. *IM*=intact enamel matrix; *NM*=necrotic enamel matrix; *DEC*=dentino-enamel cuticle; *D*=dentin.

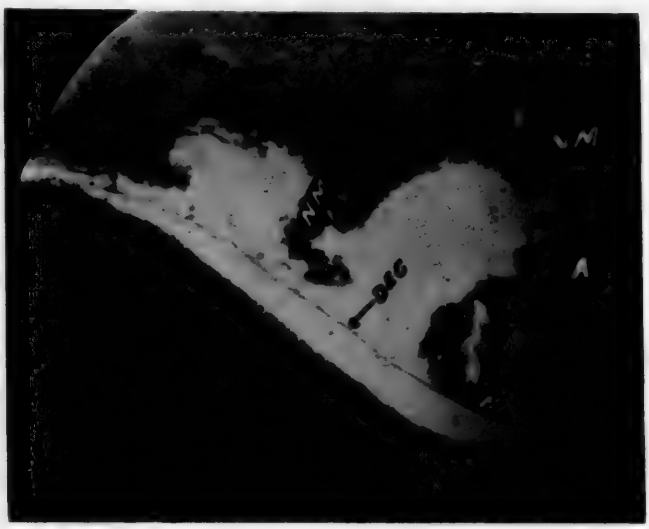

Fig. 2.—Another section of the same tooth shown in Fig. 1. Note the dentino-enamel cuticle appearing like a mere line. *IM*=intact enamel matrix; *NM*=necrotic enamel matrix; *A*=area undergoing varying degrees of necrosis of the enamel matrix; *DEC*=dentino-enamel cuticle; *D*=dentin.

seems fortunate that some portion of the enamel matrix has been detached or lost, leaving the membrane behind, otherwise it would be difficult to detect this membrane if it is in contact with an intact matrix. It may be noticed also that a portion of the dentin has been detached from the main body of the tooth and partly attached to the membrane opposite to an enamel matrix. There seems to be no doubt, therefore, that this membrane is interposed between the enamel and dentin. The writer ventures to interpret this membrane-like structure as the dentino-enamel cuticle presisting in adult human teeth.

Fig. 3.—Decalcified section of an upper molar without carious lesion. This tooth appears to have suffered a severe disturbance in dentin matrix formation. *EM*=enamel matrix; *DEC*=dentino-enamel cuticle; *D*=portion of dentin detached from the main body; *D*=dentin.

COMMENT

From the foregoing illustrations (Fig. 1 and Fig. 2) it appears that the dentino-enamel cuticle is more resistant to caries action than the enamel matrix. This observation seems to substantiate the opinion of Frisbie and Nuckolls[6] to the effect that the dentino-enamel matrix is in part responsible for the retrograde spread of caries on the enamel side before the dentin becomes infected. Our finding does not confirm the observation of Kronfeld[7] when he said: "When the dentino-enamel junction is reached, the carious process spreads much more rapidly through the less resistant dentin, thus undermining the enamel and attacking it from beneath." In our specimen, the undermining caries progresses between the dentino-enamel cuticle and the enamel matrix at the sacrifice of the latter. While portions of the enamel matrix are already necrotic and dissolved, the dentin is still intact although some dentinal tubules appear to be already affected by the carious process. The dentino-enamel cuticle seems to protect the dentin from rapid attack of caries from the enamel.

SUMMARY

1. Decalcified sections of two teeth showing a recovered enamel matrix and a membranous structure which is interpreted as the dentino-enamel cuticle

are presented in this report. From the available literature at our disposal, it seems that this is the first time that the dentino-enamel cuticle is shown present in adult human teeth.

2. The dentino-enamel cuticle appears to be more resistant to caries process than the enamel matrix. In our specimen, the undermining caries progresses between the dentino-enamel cuticle and the enamel matrix at the sacrifice of the latter. While portions of the enamel matrix are already necrotic and dissolved, the dentin appears to be still intact although some dentinal tubules seem to be already affected by the caries process.

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MERCURY ANALYSIS OF ONE HUNDRED AMALGAM RESTORATIONS

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THE physical properties of dental amalgam depend to a large degree upon the amount of mercury retained in the finished restoration. The deleterious effects of excess mercury have been well proved.[1-4] These studies have indicated that excessive mercury allowed to remain in amalgam after condensation markedly lowered the compressive strength and increased the per cent of flow. Reduction in these properties would enhance the possibilities of fracture and distortion. Other research has also shown that tarnish and corrosion occur more readily on restorations containing an excess of mercury.[5, 6, 7] These findings have led to the adoption of a technic which would expel all the mercury possible and thus assure maximum strength and resistance to tarnish. Pertinent essentials in such a technic include the use of the recommended mercury-alloy ratio, proper trituration, and correct condensation.

The recommended mercury ratio, usually 60 per cent mercury by weight, always calls for more mercury than is required in the finished restoration. This excess is added to make amalgamation easier and to obtain proper adaptation to the cavity preparation. The extra mercury is then eliminated by expressing from the semiliquid mass before condensation and by the packing procedure. The success of the restoration is dependent to a great degree upon the ability of the operator to hold to a minimum the final mercury content.

Some investigations have been done to determine the actual per cent of mercury remaining in specimens when various trituration and packing technics were employed.[4, 8, 9] These studies indicated that the final mercury content might vary from approximately 40 per cent up to 54 per cent, depending upon the manipulative procedure. Since these investigations were conducted on laboratory specimens, the actual mercury percentages might not be comparable to those present in clinical restorations. Souder and Sweeney analyzed two restorations which had been in service for over ten years and found the mercury content to be 51.6 per cent and 52.9 per cent.[10] This study also indicated that no significant amount of mercury is lost from the restoration due to slow vaporization or dissolution.

However, no one has investigated the residual mercury content in a large number of amalgam restorations. Such data would be of value not only for determining actual mercury content in the average restoration but also for detecting fluctuations which might occur in the use of various manipulative technics.

The purpose of this study, therefore, was to determine the mercury content in one hundred amalgam fillings which had been removed from extracted teeth. These teeth were secured from the Oral Surgery Department of Indiana University School of Dentistry. There was, of course, no information available in

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regard to length of service, type of alloy used, packing procedure, etc. Such data were not pertinent to this study since the primary purpose was merely to determine the residual mercury content in a typical cross section.

PROCEDURE

After the filling had been removed from the tooth, it was ground into powder by means of dental burs and stones. The powder was then sieved and only those particles which passed through a 100 mesh were used for the mercury analysis. Whenever sufficient amalgam was available, the powdered material was divided into two or three portions, making possible duplicate analyses for each restoration.

The sample of powdered amalgam was weighed on an analytical balance and then placed, by means of a charcoal boat, into a specially prepared Pyrex glass tube. This tube was medium walled, 40 cm. in length, 10 mm. in internal diameter and was sealed at one end. The tube and contents were placed in a horizontal muffle furnace and allowed to remain for 45 minutes at 700° C., with the open end of the tube extending 20 to 30 cm. beyond the end of the furnace. The mercury in the sample volitalized and condensed in this end of the tube. A cold, wet rag was wrapped around the open end to assure complete vapor condensation. After the tube had cooled down, the mercury was washed into an Erlenmeyer flask and dissolved in warm nitric acid (2:1).

The procedure for analyzing for the percentage of mercury followed the volumetric titration with thiocyanate as outlined by Hillenbrand and Lundell.[11] Tests made on known samples of pure mercury proved the method to be accurate to .02 per cent.

RESULTS

A total of 100 amalgam restorations was used and generally the mercury content of each restoration was calculated as the average of at least two separate analyses. The average residual mercury for the 100 fillings was found to be 45.4 per cent, with a minimum content of 28.6 per cent and a maximum of 61.0 per cent.

The distribution of the analyses is shown in Fig. 1. Although approximately half the fillings had a mercury content between 40 per cent and 50 per cent, there were many above and below that figure.

At the start of this investigation, it was thought that there might be a significant difference in the mercury content of various types of restorations, that is, a three-surface filling, due to the greater bulk of material, might contain more residual mercury than a one-surface restoration. However, the final tabulations show that there is no difference between the various types. Twenty-two of the restorations were one-surface amalgams and had an average mercury content of 45.5 per cent. The average in 51 two-surface restorations was 45.6 per cent, while the 27 three-surface fillings had an average residual mercury content of 45.2 per cent. It thus appears that the particular type of preparation, per se, does not markedly alter the final mercury content.

No attempt has been made to correlate the amount of residual mercury with the general condition of the restoration, such as tarnish, margins, fractured areas, etc. Without knowledge of the technic employed at the time of insertion, such a correlation would be invalid.

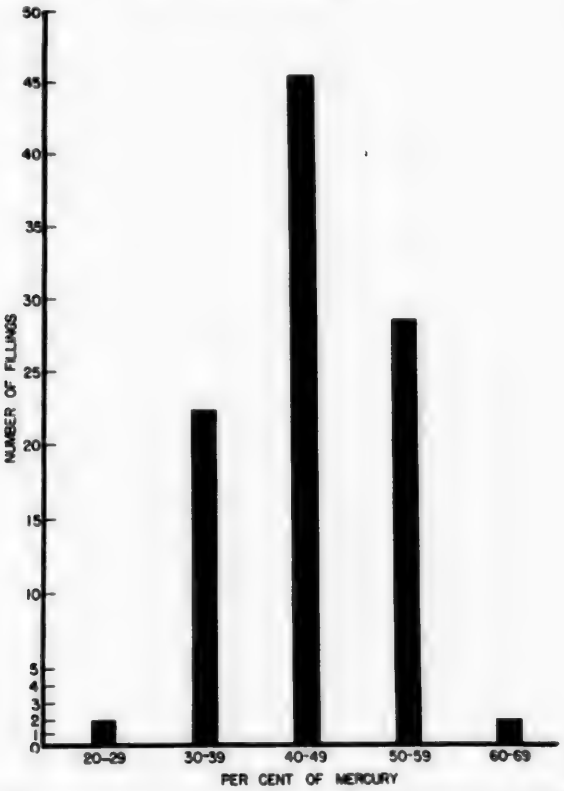

Fig. 1.—Distribution of mercury content in 100 amalgam restorations.

CONCLUSIONS

This analysis of 100 amalgam restorations shows the average mercury content to be 45.4 per cent with extremes of 28.6 per cent and 61.0 per cent. It can be seen from this research that there is a wide variation in the residual mercury content of individual restorations, dependent upon the manipulative technic used. Attention to certain fundamentals can minimize the amount of mercury remaining in the restorations and thus assure maximum strength and resistance to tarnish.

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SUGGESTED REVISIONS FOR TESTING DENTAL ELASTIC IMPRESSION MATERIALS

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IN the development and plant control of elastic dental impression materials, of both the agar and alginate base types, it is necessary to rate materials as to their respective efficiency for practical use in order to select the best formulation or manufacturing process. The tests most commonly used for this purpose are those outlined in A. D. A. Specification #11 for hydrocolloidal impression materials[1] and the Medical Department Tentative Specification No. MD-1285D for alginate impression materials.

Rating the efficiency of experimental impression materials from the data obtained in these tests does not always check the experience of operators in clinical tests. Therefore this study of measurement of the various physical properties of elastic impression compounds was instituted after preliminary investigation had indicated that the specification tests could be altered, somewhat, to permit a more accurate comparison and a better evaluation of the efficiency of the impression materials. Considering the properties such a material should possess, they would include: (1) ability to pass undercuts without rupturing, (2) ability to return to the original shape, and (3) rigidity sufficient to resist undue deformation when plaster or stone is poured into the impression material.

The specifications now used for the two types of material provide tests for (1) strain, or deformation caused by a given load, (2) set, or permanent deformation resulting from that strain, and (3) the compressive load required to rupture the material. These three specification tests serve as a fairly good guide in the evaluation of an impression material but do not, in every case, yield results which are comparable.

TABLE I

MATERIALS INVESTIGATED

TRADE BRAND	MANUFACTURER	ADDRESS
Hydrocolloids:		
Deelastic Hydro Colloid Impression Material	Thomas J. Dee & Co.	Chicago, Ill.
Dentocoll	The L.D. Caulk Company	Milford, Del.
D-P Impression Colloid	Dental Perfection Co.	Los Angeles, Calif.
Kerr Hydrocolloid Elastic Impression Material	Kerr Manufacturing Co.	Detroit, Mich.
S. S. White Elastic Colloid	The S.S. White Dental Mfg. Co.	Philadelphia, Pa.
Alginates:		
Coe-Loid Powder	Coe Laboratories, Inc.	Chicago, Ill.
D-P Elastic Impression Cream	Dental Perfection Co.	Los Angeles, Calif.
Getz 400 Elastic Impression Powder	The William Getz Co.	Chicago, Ill.
Reprolastic Powder	Reliance Dental Mfg. Co.	Chicago, Ill.
Zelex	The L.D. Caulk Company	Milford, Del.

As an illustration of the difficulties confronting an investigator in evaluating the results obtained from the specification tests, five brands of each type of material were examined. The brand names of these materials, together with the names of their respective manufacturers, are listed in Table I.

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The materials were first tested according to the procedures outlined in the previously mentioned specifications. In all cases, in this and all succeeding tests outlined in this paper, the materials were handled according to the enclosed manufacturers' directions. The results obtained for these specification tests are listed in Tables II and III. The code letters in these and succeeding tables are not correlated with the materials as listed in Table I.

TABLE II

SPECIFICATION TEST RESULTS OF HYDROCOLLOIDS

HYDROCOLLOID	GEL TEMPERATURE (°C.)	STRAIN (%)	SET (%)	STRENGTH (GM./CM.²)
A	37	4.6	0.5	2770
B	36	8.8	1.0	2370
C	35	14.7	2.2	3170
D	37	9.5	1.3	1830
E	35	2.9	0.4	2500
Specification	35-45	4-20	3.0 maximum	2000 minimum

TABLE III

SPECIFICATION TEST RESULTS OF ALGINATES

ALGINATE	SETTING TIME (MINUTES)	STRAIN (%)	SET (%)	STRENGTH (GM./CM.²)
A	5	16.6	3.2	2900
B	3½	13.2	2.7	3030
C	4	11.3	2.9	3470
D	3	8.2	2.0	3800+
E	3	6.8	2.0	3800+
Specification	6 ± 2	4-20	6.0 max.	2000 minimum

Comparison with the specification requirements shows Hydrocolloids D and E unsatisfactory, D failing to meet the required minimum strength of 2000 Gm./cm.² and E exhibiting insufficient strain. All of the alginates tested pass the specification requirments for strain, set, and strength but B, D, and E all gel before the minimum four minutes required.

Having obtained these results, it should be possible to evaluate properly the relative merits of each material with respect to each of the properties tested. If the pertinent properties have been tested, and if it has been done in such a way as to permit comparison between the various materials, then the relative worths of them as impression materials can be determined.

Considering the properties of strain, strength, and set in the order named, that of strain would seem to be a valuable property to determine for it gives a measure of the rigidity of the material. Also, since the same load is applied to each material, the results obtained for the ten materials can be compared and the relative rigidities of the materials determined. The compressive strength values are also comparable but, as will be pointed out later, can lead to misleading conclusions.

The data for set are subject to difference in opinion as to their interpretation. By one course of reasoning the lowest value for set should indicate the best material, for it indicates a smaller amount of permanent deformation in the impression when drawn over an undercut, and hence a more accurate working cast. Accordingly, the hydrocolloids would be rated as E best; with A, B, D, C following in that order and the alginates would be rated as E, D, B, C, A with only D and E better than the poorest hydrocolloid, C.

This is the type reasoning employed by Skinner and Pomés[2] who describe their method of obtaining set values by applying stresses of 27.6, 400, 0, and 27.6 Gm./cm.^{2} for intervals of ½, 1, 2, and 2½ minutes, respectively. Employing this procedure, they determined set values for eight different alginates at various times after gelation. In discussing their results, they state, "Not only must the impression be strong enough not to fracture on withdrawal, but also it must be sufficiently rigid and possess the proper elasticity to return closely to its original shape after it has been withdrawn over undercuts. Consequently, the lower the amount of permanent set values shown . . . the more satisfactory is the material."

This interpretation of the test data may not be sound, however, since it must be realized that the set values are derived from varying amounts of strain; and that the resultant set is directly dependent upon the amount of strain to which the material is subjected.

EXPERIMENTAL BACKGROUND FOR PROPOSED TEST

In order to determine the relationship of strain and set of the ten materials investigated, a series of tests was run on each material. Six test cylinders 0.5 inch in diameter and 0.75 inch high, the same as used in specification tests, were prepared from each stick of hydrocolloid or mix of alginate. These were permitted to age for one hour, being stored in 100 per cent relative humidity during the final one-half hour before starting the test, and the following set of tests was then run on each material.

Using the specification testing apparatus, to the first cylinder a load of 100 Gm./cm.^{2} was applied, and at the end of thirty seconds a reading of the cylinder height taken by means of a dial gauge (Reading A). At the end of the first minute an additional deforming load of 400 Gm./cm.^{2} was applied to the cylinder, and at the end of thirty seconds a second reading of the dial gauge was taken (Reading B). At the completion of the second minute, all load was removed for one minute. The original load of 100 Gm./cm.^{2} was then reapplied, and thirty seconds later a third dial gauge reading taken (Reading C). As set forth in the specification, the difference between readings A and B divided by the original length of the specimen times 100 was considered the percentage of strain and the difference between readings A and C divided by the original length times 100 was considered the percentage of set. This same test procedure was followed with the remaining five cylinders except that in each case successively larger deforming loads were applied.

Since the amount of strain produced in the cylinder is dependent upon the amount of stress applied to it, by this test procedure a series of values for strain were obtained, and at the same time the sets caused by those strains were determined.

Three series of tests were run on each material investigated and the 18 values for strain and set were plotted against each other. Through these points was drawn a strain-set curve for each material. In Figs. 1 and 2 are the curves obtained for the hydrocolloids and alginates, respectively. These curves permit a ready comparison of the relative amounts of set to be expected from each at a particular strain or, from a practical standpoint, when drawn over the same undercut.

Since the material exhibiting the least set for a given amount of strain will most accurately reproduce undercuts, the materials would be rated as shown in Tables IV and V. Also shown is the previously drawn rating derived from the specification test results.

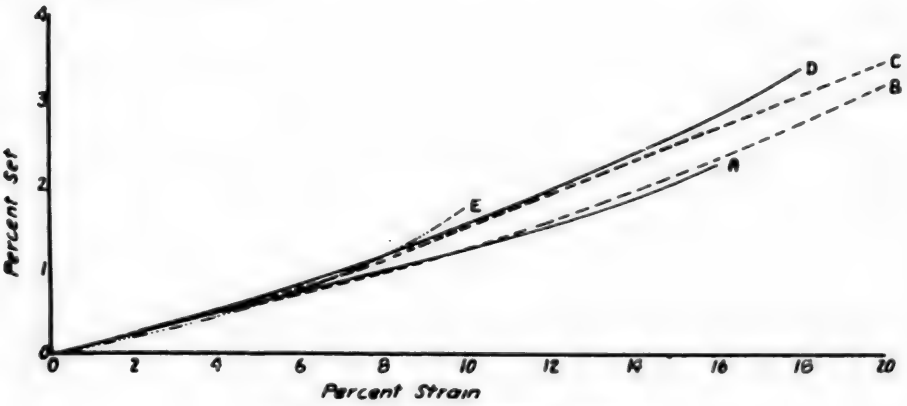

Fig. 1.—Agar base impression materials.

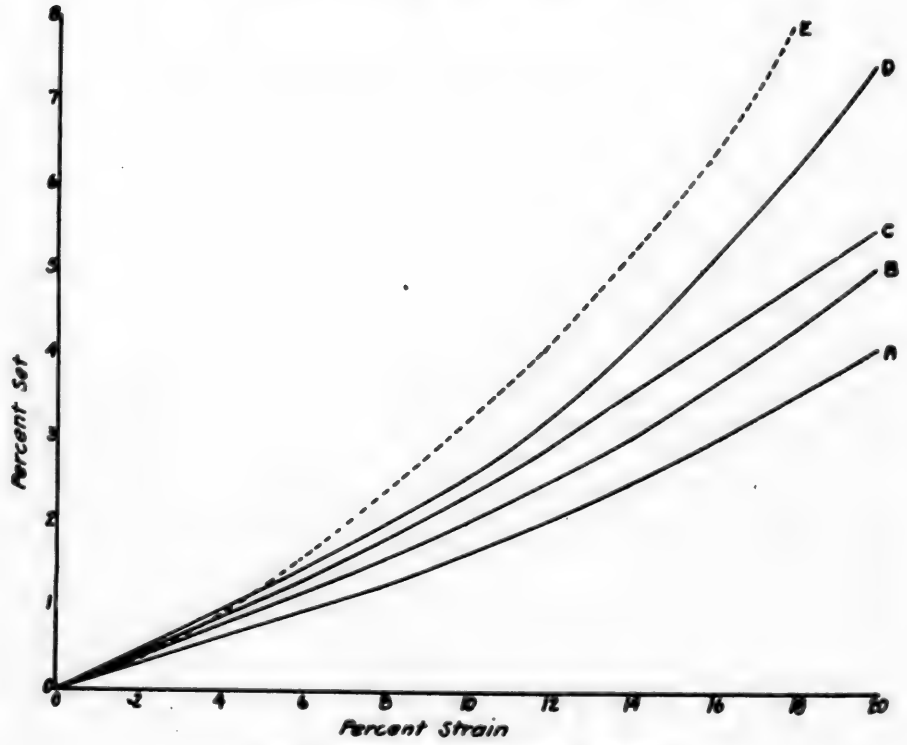

Fig. 2.—Alginate base impression materials.

TABLE IV

HYDROCOLLOIDS RATED ACCORDING TO INCREASING SET

SPECIFICATION TEST	STRAIN-SET CURVES
E	A
A	B
B	C
D	D
C	E

TABLE V

ALGINATES RATED ACCORDING TO INCREASING SET

SPECIFICATION TEST	STRAIN-SET CURVES
E	A
D	B
B	C
C	D
A	E

It is apparent that the two sets of ratings are quite different and since the primary object is to ascertain how well the materials will regain their original dimensions when withdrawn over any particular undercut rather than when subjected to the same compressive force, the ratings drawn from the specification test would seem to be misleading.

Such strain-set curves supply valuable information, but obtaining the data required is rather time consuming and tedious. It was therefore felt necessary to devise some simpler means of accomplishing the same result.

PROPOSED TEST

Since the relative positions of the curves do not change with respect to one another over the range investigated, with the sole exception of Hydrocolloid E whose slope of curve increases rapidly as it approaches its rupture point, it was felt that the determination of set at an arbitrarily chosen strain would serve to indicate the relative degrees of set of the various materials. Since the specifications require the strain to be between 4 per cent and 20 per cent the mid-point, or 12 per cent, was selected as the strain to be applied to the cylinders. To attempt to obtain exactly 12 per cent strain by finding the exact amount of weight required would be most impracticable so this method was not followed.

Several alternative methods were tried, the most obvious one being the use of a ring "stop" cut to 88 per cent the height of the cylinder molds. This, however, was found to be unsatisfactory since, for materials of different rigidities, the initial strains caused by applying a light load were different and hence the amount of additional strain produced in compressing them to the level of the stop was not constant. Eventually a slightly different type of testing apparatus was developed (Fig. 3).

The procedure followed in testing with this instrument consisted of lowering the dial gauge, which exerted a positive pressure of 40 Gm./cm.^{2}, onto the cylinder and thirty seconds later reading the dial gauge (Reading A). From this reading, 2.28 (the distance in mm. required to produce 12 per cent strain) was subtracted and at the one minute point the cylinder was compressed to this reading by means of the screw mounted above the dial gauge. This com-

pression was maintained for one minute after which all load was removed from the cylinder for one minute. The dial gauge was then again lowered onto the cylinder, and thirty seconds later a second reading taken (Reading B). The difference between Readings A and B divided by the original length of the cylinder times 100 was again considered the set in percentage.

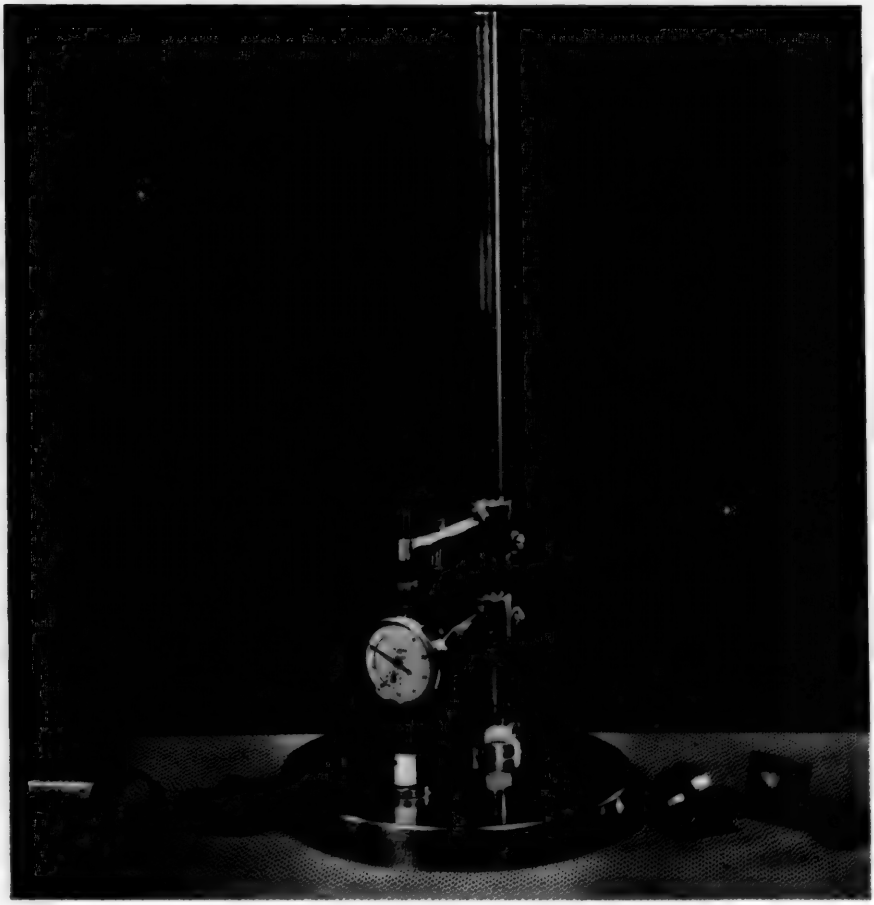

Fig. 3.—Test apparatus used to obtain 12 per cent strain.

At least three cylinders of each material were so tested, the average results obtained being tabulated in Tables VI and VII, together with the value for set at 12 per cent strain taken from the curves appearing in Figs. 1 and 2.

TABLE VI

HYDROCOLLOIDS—PER CENT SET AT 12 PER CENT STRAIN

HYDROCOLLOID	STRAIN-SET CURVE	12% STRAIN TEST
A	1.5	1.6
B	1.6	1.5
C	1.9	1.7
D	2.0	1.8
E	Broke	Broke

TABLE VII

ALGINATES—PER CENT SET AT 12 PER CENT STRAIN

ALGINATE	STRAIN-SET CURVE	12% STRAIN TEST
A	2.1	2.2
B	2.5	2.6
C	2.9	2.8
D	3.2	3.5
E	4.1	3.7

Typical data for the "set at 12 per cent strain" test showing the spread on duplicate specimens is shown for Hydrocolloid A and Alginate A in Table VIII.

TABLE VIII

TYPICAL DATA ON SET AT 12 PER CENT STRAIN TEST

MATERIAL	STRAIN (%)	SET (%)
Hydrocolloid A:		
Cylinder 1	12	1.63
Cylinder 2	12	1.52
Cylinder 3	12	1.52
Average		1.56
Alginate A:		
Cylinder 1	12	2.32
Cylinder 2	12	2.06
Cylinder 3	12	2.16
Average		2.18

The results obtained from this test are in quite good agreement with those found on the strain-set curves and indicate that this method of testing can be applied to gaining a measure of the relative sets of the various materials when pulled over the same undercut. In addition to yielding results in accord with those on the curves, it is much less cumbersome, requires less time, and calls for no elaborate testing apparatus.

As has been stated earlier, these tests were conducted after the cylinders had aged for an hour. The specifications for both hydrocolloids and alginates call for this treatment and it was adhered to in the foregoing tests.

In the case of the alginates, however, where the physical properties of the material depend upon the formation of an insoluble alginate gel by a chemical reaction, it was felt that tests run one hour after the initial gelation had taken place might well yield results not indicative of the properties of the materials at the time they are subjected to deformation. The formation of insoluble alginate gel continues for some time after the initial gelation occurs, resulting in a corresponding development of physical properties, and since the primary objective is to determine the ability of the material to pass over undercuts and then return to its original shape at the time an impression would normally be removed from the mouth, tests conducted at such a time would be more illustrative of the performance to be expected.

As a test for the resistance to permanent deformation developed in a practicable time, the test for set resulting from 12 per cent strain was repeated on three cylinders of each material. This time, however, the test was started on the first cylinder two minutes after gelation had taken place. The remaining two

cylinders were tested at four-minute intervals, the time required to complete a test and change the cylinders. The results shown in Table IX are the averages of three such runs.

TABLE IX

ALGINATES

TIME FROM GELATION IN MINUTES	PER CENT SET AT 12 PER CENT STRAIN				
	A	B	C	D	E
2	2.6	3.3	4.1	3.6	3.8
6	2.2	3.1	3.4	3.6	3.4
10	2.3	2.6	3.4	3.6	3.6

The two-minute values are doubtlessly the most important since it is in this range of time that most impressions are withdrawn from the mouth. In the case of Materials A, B, and C the set values at two minutes are found to be appreciably higher than those found at one hour, C exhibiting the slowest development of resistance to permanent deformation. Materials D and E are found to gain rapidly almost their ultimate resistance to permanent deformation but still yield higher sets than A and B. This test indicates that, at the time the materials would normally be subjected to deformation, the accuracy of reproduction to be expected, in order of decreasing accuracy, would be A, B, D, E, and C.

Since Material C is found to be slower in gaining resistance to permanent deformation than are the rest, and since Materials A and B likewise exhibit higher set values several minutes after setting than they do after an hour, the indications are that such a test should be run a few minutes after the gelation time in order to obtain the best data for this property.

Two-minute tests for strain and strength were also made on each alginate. The strain test was started two minutes after gelation had taken place and was conducted in the same manner as the specification test, applying 100 and 1000 Gm./cm.^{2} loads to the cylinder. New cylinders were prepared for the strength test so that it too could be started two minutes after gelation. At the two-minute mark a load of 500 Gm./cm.^{2} was applied to the cylinder and each thirty seconds thereafter an additional load of 400 Gm./cm.^{2} was applied until the cylinder ruptured. The results of these two tests are shown in Table X, together with the corresponding values obtained in the one-hour specification tests, shown in Table III.

TABLE X

STRAIN AND STRENGTH OF ALGINATES TWO MINUTES AND ONE HOUR AFTER GELATION

ALGINATE	TWO MINUTES AFTER GELATION		ONE HOUR AFTER GELATION	
	STRAIN (%)	STRENGTH (GM./CM.^{2})	STRAIN (%)	STRENGTH (GM./CM.^{2})
A	21.2	1300	16.6	2900
B	13.5	2900	13.2	3030
C	14.2	2500	11.3	3470
D	12.7	3800+	8.2	3800+
E	14.4	3800+	6.8	3800+

Comparing the two-minute and one-hour values in Table X, Materials A, C, D, and E are found to possess higher strain values two minutes after gelation than at one hour after gelation. This indicates that these materials have not, at

the two-minute point, developed their ultimate degree of rigidity. Material B, however, shows a rapid development of rigidity having virtually the same strain value at two minutes as it does in one hour. All the materials, with the exception of A, show a rapid development of strength.

Concurrently with the strength test just outlined, the amount of strain which had occurred when the material reached its rupture point was determined. This was done by observing the dial gauge reading prior to the application of the initial load and the reading at the point of rupture. The difference between the two readings, expressed in percentage of original length, was considered the amount the material is capable of being deformed without rupturing. This same type test was also run on the hydrocolloids after aging an hour. These results are shown in Tables XI and XII.

TABLE XI

PER CENT STRAIN AT POINT OF RUPTURE OF HYDROCOLLOIDS AGED ONE HOUR

HYDROCOLLOID	STRAIN AT RUPTURE (%)
A	23
B	25
C	50
D	22
E	12

TABLE XII

PER CENT STRAIN AT POINT OF RUPTURE OF ALGINATES TWO MINUTES AFTER GELATION

ALGINATE	STRAIN AT RUPTURE (%)
A	25
B	30
C	25
D	Greater than 50
E	40

The results shown in Tables XI and XII are of interest when considered together with the compressive strength values shown in Tables II and X. Since Alginate A does exhibit a much lower strength than the rest, it might be considered that it would prove inferior because of the likelihood of its tearing or breaking more readily. The results appearing in Table XII, however, show that Alginate A is capable of being deformed 25 per cent of its length before rupturing. This figure is identical with that of Alginate C which has a strength almost twice as great. Hydrocolloid C is the only one of that type which can withstand more deformation than Alginate A. Hydrocolloid E which shows a strength of 2500 Gm./cm.^{2} still can be deformed only about 12 per cent before breaking, or just about half as much as Alginate A which has a strength of only 1300 Gm./cm.^{2}. The data, therefore, shows that the amount of deformation materials of these types can withstand without failure is not dependent on their high strength!

CONCLUSIONS

The tests for physical properties of dental elastic impression materials included in the A. D. A. Specification No. 11 for hydrocolloidal impression ma-

terials and the Medical Department Tentative Specification No. MD-1285D for alginate impression materials measure: (1) strain, or deformation caused by a given load, (2) set, or permanent deformation resulting from that strain, and (3) the compressive load required to rupture the material.

This investigation indicates that these specification tests do not truly measure the value of such a material and yield results which are open to misinterpretation.

The values for "set" as determined by specification methods do not permit comparison of the relative abilities of the materials to return to their original dimensions after being drawn over any given undercut since they result from varying amounts of "strain" that ensue from the application of a constant deforming load to materials of varying rigidities.

Determination of set from a constant amount of strain yields values which are comparable.

The amount of deformation materials of these types can withstand is not dependent on their high strength. Strain at point of rupture more truly measures the ability of materials to resist rupture than does compressive strength.

The properties required of a dental elastic impression material include: (1) ability to pass over undercuts without rupturing, (2) ability to return to the original dimensions after passing undercuts, and (3) rigidity sufficient to resist undue deformation when plaster or stone is poured into the impression.

These properties can be readily measured by determining: (1) strain at point of rupture, (2) set at 12 per cent strain, and (3) strain caused by a constant load.

Since impressions are removed from the mouth several minutes after gelation occurs, and since the development of physical properties will continue well beyond that time in the case of alginates, "set at 12 per cent strain" and "strain at point of rupture" of alginates should be determined several minutes after gelation.

Actual impression taking, as of the undercut posts standard model described in the present specifications, is a very pertinent test for it parallels the manner in which impressions are handled in practical usage. Paffenbarger[1] has discussed the effect of the time factor of applying strain upon rupture and shown that a material will withstand a much greater strain if it is applied and released rapidly. Such a procedure can best be followed by actually taking an impression of such a standard model.

SUMMARY

The physical properties of elastic impression materials as determined by specification tests do not truly measure the value of such a material and are subject to misinterpretation.

A new series of tests have been designed to more truly determine the pertinent properties and permit direct comparison of materials.

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1. Paffenbarger, G. C: Hydrocolloidal Impression Materials: Physical Properties and a Specification, *J. A. D. A.* **27**: 373, 1940.
2. Skinner, E. W., and Pomés, C. E.: Alginate Impression Materials: Technic for Manipulation and Criteria for Selection, *J. A. D. A.* **35**: 245, 1947.

THE HYDROGEN-ION CONCENTRATION OF THE SALIVA

II. The Relationship Between Hydrogen-Ion Concentration and Rate of Flow of Saliva

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THE acidity of the saliva is an aspect of salivary secretion which has received considerable attention, chiefly because of the intimate relationship between the saliva and teeth and the possible connection between dental disease and salivary composition. The saliva is of more general interest, however, as an example of secretion produced by a compound secretory epithelium; and saliva is available for study uncontaminated to any great extent by other secretions. One important aspect of any secretory process is the effect of changes in the composition of the blood transmitted to the glands via the tissue fluid upon the secretion produced by the glands.

The experiments to be described were designed, first, to establish a method by which measurements of the $[H^+]$ of the saliva could be made with a high degree of accuracy, and, second, to investigate the effect on salivary $[H^+]$ of substances known to alter the alkali reserve of the blood plasma.

METHOD

The Stadie microglass electrode for use with the Cambridge Instrument Co. pH meter was chosen for pH determinations because its small size would make possible direct measurements in the mouth. The method of intraoral measurement of salivary pH offered three distinct advantages: (1) no precautions against CO_2 loss were necessary, provided the mouth was kept closed while the electrode was in position; (2) measurements could be made over a period of five or ten minutes, or longer if required, without removing the electrode from the mouth, and (3) all measurements would be strictly comparable, since they were made at mouth temperature.

The electrode system is shown in Fig. 1. Contact between the saliva and the calomel reference electrode was made by a small capillary tube connected by a flexible tube to a beaker containing saturated KCl. The capillary and flexible connecting tube contained agar-agar jelly made up with saturated KCl. The calomel electrode dipped into a beaker of saturated KCl, thus completing the electrical circuit. It was found necessary, for steady readings, to shield the electrode from contact with the tongue and mucous membrane. This was achieved by the glass sleeve shown in the diagram. The lower end of the sleeve was open and curled round on itself and allowed saliva to enter. A constant flow of saliva past the electrode system was maintained by including in the side arm of the sleeve a narrow bore gutta-percha catheter tube. The end of this

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hung down outside the mouth forming a syphon. The syphon flow was slower than even the minimum salivary secretion, and it was always necessary to augment the flow at intervals by withdrawing saliva with a syringe attached to the catheter tube.

pH measurements were made as follows: Saliva was allowed to collect in the mouth for one minute and, at the end of this time, transferred by pressure of the lips to the electrode sleeve. The electrode system was then placed in position in the mouth with its curled end under the tongue in a pool of saliva and its cable held firmly between the lips. Syphoning was started and readings were taken at one-minute intervals. It was found that the first five readings tended to show some fluctuation, but the second five seldom varied by more than 0.02 of a pH unit.

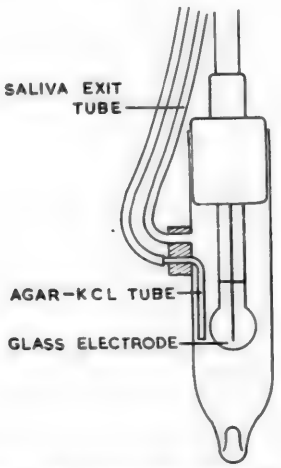

Fig. 1.—Electrode system for direct measurement of saliva pH in the mouth.

The readings were converted to [H^+] and the mean of the second five values was taken as the prevailing [H^+] of the saliva at that time. The time was noted as soon as saliva collection started, and all the saliva which syphoned out of the mouth, as well as that remaining at the end of the measurement period, was collected and its volume recorded. In this way it was possible to determine the rate of flow of saliva. Potassium hydrogen phthalate at 37° C. with a pH of 3.97 was used for standardization of the glass electrode before every series of measurements.

With the object of ascertaining the diurnal variations, records were taken six times during the day for a period of two weeks. The times chosen were the following: Three hours after breakfast (10:30 A.M.), before lunch (12:45 P.M.), immediately after lunch (2:00 P.M.), one hour after lunch (3:00 P.M.), before tea (3:45 P.M.), and immediately after tea (4:15 P.M.).

A second series of experiments was planned to determine the effect on salivary [H^+] of substances known to alter the plasma alkali reserve. Since no significant variation in salivary [H^+] was found between 10:30 A.M. and 3:00

P.M., this period was taken for the alkali reserve experiments, NH_4Cl in 5-Gm. doses being chosen as the acidifying agent and $NaHCO_3$ in 10-Gm. doses as the alkalinizing agent. The NH_4Cl brought about a decrease of the order of 40 mg. and the $NaHCO_3$ an increase of approximately 35 mg. in the venous plasma bicarbonate content as estimated with the Van Slyke volumetric apparatus. The experiments were planned in a three-day rota, made up of one NH_4Cl, one $NaHCO_3$, and one control day. The sequence of events in the control day was identical with the other two days except that the dose was 350 ml. of water alone. The days were set out in random order to minimize the possibility of cumulative effects of the drugs.

The daily sequence was as follows:

10:30 A.M	Saliva pH and flow recorded, sample of venous blood taken for plasma bicarbonate determination.
10:45 A.M	Dose taken (NH_4Cl, or $NaHCO_3$, or water control).
11:30 A.M.	Saliva pH and flow recorded.
11:45 A.M.	Venous blood sample taken.
12 noon	Saliva pH and flow recorded.
12:30 P.M.	Saliva pH and flow recorded.
12:45 P.M.	Venous blood sample taken.
1:00 P.M.	Lunch.
2:15 P.M.	Saliva pH and flow recorded.

RESULTS

Table I shows the mean $[H^+]$ for the six daily measurements during the control period of twelve days. In order to follow any trends throughout the day, the differences between the means were tested using Student's "t" distribution, taking a probability of 0.05 as the level of significance. The results obtained before and after meals were taken altogether and tested, and a comparison was also made between the mean $[H^+]$ at 10:30 A.M. and 4:15 P.M. The results of these tests are shown in Table I, and it is seen that the only significant difference in $[H^+]$ is between 3:00 P.M. and 3:45 P.M. and between 10:30 A.M. and 4:15 P.M.

The daily mean $[H^+]$ and rate of flow are given in Table II. Saliva flow was only recorded for the last nine days, and on inspection of the results it is

TABLE I

SHOWING THE MEAN $[H^+]$ OF THE SALIVA AT DIFFERENT TIMES DURING THE DAY AND THE RESULTS OF STATISTICAL TESTS TO ESTIMATE THE SIGNIFICANCE OF THE DIFFERENCES BETWEEN THE MEANS

TIME	MEAN $[H^+]$ $\times 10^{-7}$ GM. IONS/LITER	MEAN DIFFERENCE	σ DIFFERENCE	"t"	P
10:30 A.M.	1.999	0.322	± 0.2078	1.55	> 0.1
12:45 P.M.	1.677				
2:00 P.M.	1.554	0.250	± 0.1520	1.64	> 0.1
3:00 P.M.	1.804	0.173	± 0.0762	2.27	< 0.05
3:45 P.M.	1.629				
Before meals	1.622	0.093	± 0.0705	1.31	> 0.2
After meals	1.529				
10:30 A.M.	1.999	0.424	± 0.1346	2.48	< 0.05
4:15 P.M.	1.575				

seen that the [H⁺] tends to rise in the latter half of the period while the rate of flow falls. The gradual reduction in flow probably accompanied the development of a tolerance towards the presence of the apparatus in the mouth. The apparent relationship between [H⁺] and rate of flow became clearer when the individual readings were examined, and it was therefore decided to prepare a scatter diagram of all the control results, with a view to investigating a possible correlation. The diagram is shown in Fig. 2. The correlation coefficient was calculated to be -0.708 ± 0.1066, showing a fairly high degree of negative correlation between [H⁺] and rate of flow. The nonlinearity of the regression lines in the region of low rates of flow may possibly be due to bacterial acid production becoming a significant factor in salivary [H⁺] at this level.

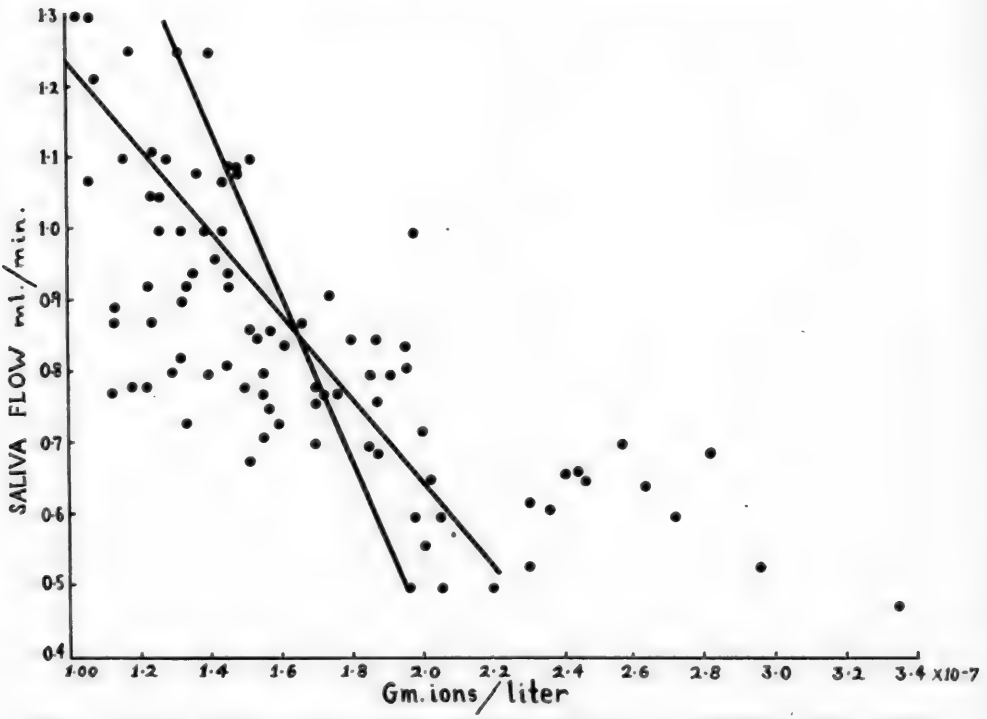

Fig. 2.—Scatter diagram showing saliva flow and hydrogen-ion concentration during control periods.

The relationship between [H⁺] and rate of flow had to be considered when interpreting results obtained after ingestion of acidifying or alkalinizing drugs. These drugs might have an effect on saliva flow with secondary alterations in [H⁺] apart from any effect on [H⁺] brought about as a direct result of their acidifying or alkalinizing properties. In fact, NH_4Cl did produce an increase in salivation, especially on the occasions when nausea was experienced. If this were its only effect, then a fall in the saliva [H⁺] was to be expected.

TABLE II

SHOWING DAILY MEAN [H^+] AND STANDARD ERROR DURING THE TWELVE-DAY PERIOD AND THE DAILY MEAN RATE OF FLOW IN THE LATTER NINE DAYS OF THIS PERIOD

DAY	MEAN [H^+] $\times 10^{-7}$ GM. IONS/LITER	RATE OF FLOW ML./MIN.
1	1.523 ± 0.216	
2	1.211 ± 0.330	
3	1.216 ± 0.204	
4	1.479 ± 0.389	0.84
5	1.280 ± 0.116	0.91
6	1.469 ± 0.258	0.91
7	1.547 ± 0.259	0.82
8	1.460 ± 0.132	0.79
9	2.034 ± 0.853	0.71
10	1.970 ± 0.419	0.71
11	2.596 ± 0.547	0.62
12	2.382 ± 0.502	0.59

Scatter diagrams of [H^+] and rate of flow after $NaHCO_3$ and NH_4Cl were prepared, and again a negative correlation between [H^+] and flow were shown though not so great as in the control periods. The correlation coefficient after $NaHCO_3$ was -0.494 ± 0.218 and after NH_4Cl -0.656 ± 0.186. Table III gives the mean [H^+] and rate of flow during the control, NH_4Cl, and $NaHCO_3$ periods. The differences were tested statistically and the results of the tests are shown in the table. All the differences reached the level of significance required, except the difference in rate of flow between the control and $NaHCO_3$ periods.

TABLE III

SHOWING THE MEAN [H^+] AND RATE OF FLOW OF SALIVA IN THE CONTROL PERIOD AND FOLLOWING NH_4Cl AND $NaHCO_3$. THE RESULTS OF STATISTICAL TESTS TO ESTIMATE THE SIGNIFICANCE OF THE DIFFERENCE BETWEEN THE MEANS ARE SHOWN

	MEAN [H^+] $\times 10^{-7}$ GM. IONS/LITER	DIFFERENCE	"t"	P
NH_4Cl	1.87			
		0.22	2.754	< 0.01
Control	1.65			
		0.24	2.897	< 0.01
$NaHCO_3$	1.42			
	MEAN FLOW ML./MIN.	DIFFERENCE	"t"	P
NH_4Cl	0.933			
		0.086	2.783	< 0.02
Control	0.847			
		0.004	0.913	> 0.3
$NaHCO_3$	0.813			

DISCUSSION

The results reported have been obtained over a long period of time at frequent intervals during the day and show the remarkable constancy of the salivary [H^+]. This confirms the results of Grossman and Brickman (1937), Eisenbrandt (1943), and Schmidt Nielsen (1946). Meals appear to have no effect on the salivary [H^+] measured immediately before and after eating. The statistically significant fall in [H^+] between 3:00 P.M. and 3:45 P.M. may be a reflection of the so-called "alkaline tide" changes in the plasma alkali reserve, but no records of the latter were taken in the first series of experiments. In terms of pH, the range of values obtained in the entire experimental period was between 6.46 and 7.00. Within this range there appears to be some negative

correlation between [H^+] and rate of flow, which suggests that the saliva [H^+] approaches more nearly to that of the plasma as the secretion rate increases. However, no figures are available with higher rates of flow, but the work of others and results obtained in class experiments show that the saliva can become more alkaline than the plasma.

The results in the second series of experiments demonstrate that the effects of NH_4Cl and $NaHCO_3$ on the plasma were reflected in the saliva even though the relationship between [H^+] and flow established in the control experiments was maintained. Benetato and Oprisiu (1935) reported similar changes, and went so far as to postulate that the salivary glands were playing a part in acid-base regulation. Such a role could only be of very small importance compared with the kidney, even if the ultimate fate of hydrogen ions in the saliva were certain. The mean hydrogen-ion concentration for the control period, expressed in pH units, was 6.783. After NH_4Cl it fell to 6.728 and rose to 6.848 after $NaHCO_3$. Thus, accompanying quite large changes in the alkali reserve of the blood, the mean salivary pH was altered by about 0.06 of a unit, a measurement only just outside the experimental error of the method. The clinical importance of such a change must be negligible.

In assessing the value of the results, it must be emphasized that they were obtained on one subject only, and it is possible that acidifying and alkalinizing drugs might have a more profound effect in other subjects. The pH changes recorded, however, are of the same order as the concomitant changes in the blood under similar conditions, and do not suggest that any special acid-base regulating activity is possessed by the salivary glands.

SUMMARY

1. A method of intraoral measurement of salivary pH is described.

2. Results obtained show a negative correlation between [H^+] and rate of flow of saliva.

3. A statistically significant change in saliva [H^+] has been produced by alkalinizing and acidifying drugs.

4. The physiological and clinical significance of these changes is discussed.

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THE RELATION BETWEEN BUFFERING CAPACITY, VISCOSITY, AND LACTOBACILLUS COUNT OF SALIVA

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THE possible relationship between the buffering capacity, neutralizing power, titratable alkalinity of saliva, and the prevalence of dental caries has engaged the attention of various workers for the past 35 years.

Marshall[1] devised a "salivary factor" as an index of caries activity which he found to be fairly constant for an individual. It was the quotient of the neutralizing power of "resting saliva" divided by the neutralizing power of paraffin-stimulated saliva, multiplied by 100. In immune individuals he found that this factor varied between 80 and 132.

Shepard and Gies[2] criticized Marshall's work and published results showing that his salivary factor was neither constant for an individual nor an index of caries activity. Forbes and Gurley[3] stated that saliva from caries-immune individuals had a high acid neutralizing action whereas saliva from caries-susceptible individuals had a low acid neutralizing action. Karshan, Krasnow, and Krejci[4] stated that the titratable alkalinity of immune saliva is higher than that of susceptible saliva if stimulated saliva is used but that there is no significant difference if resting saliva is used.

Wills and Forbes[5] studied the effect of diet on the acid neutralizing power of saliva and reported that on a low carbohydrate, high protein diet an increased capacity of the saliva to neutralize acid was observed. Dreizen, Mann, Cline, and Spies[6] stated that the neutralizing power of saliva of nutritionally deficient caries-free individuals is greater than that of well-nourished individuals with rampant caries. They also stated that a high buffering capacity and a low lactobacillus acidophilus count go together. Since diet was another complicating variable in these experiments, it is hard to draw conclusions about the relation between buffering capacity and dental caries. Many of these workers measured the titratable alkalinity with the exception of Dreizen, Mann, Cline, and Spies.

A distinction must be maintained between "buffering capacity"—the ability of the saliva to resist a change in pH and the titratable alkalinity—the amount of acid required to bring the pH to the place where an arbitrarily chosen indicator changes colour. The former is best measured by the slopes of the titration curves, the latter by the ml. of acid needed to produce a pH of say 4.0.

Two examples will illustrate this difference. Saliva from T.V. required 6 ml. of 0.01M HCl to produce pH of 6.00 and saliva from L.R.P. also required 6 ml. of 0.01M HCl to produce a pH of 6.00. With litmus as an indicator, these two salivas would show the same titratable alkalinity. The slopes of the curves obtained by plotting ml. of acid used against pH are, however, widely different.

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The value from T.V. is 0.311 and from L.R.P. 0.235. The initial pH's of all the salivas (126 samples) were remarkably uniform, the over-all average being 7.55.

The term "resting saliva" and values based on this rather indefinite term seem to us to be poor criteria to correlate with dental caries if the values for stimulated salivas show no such correlation, and vice versa. For this reason stimulated salivas were used throughout this experiment.

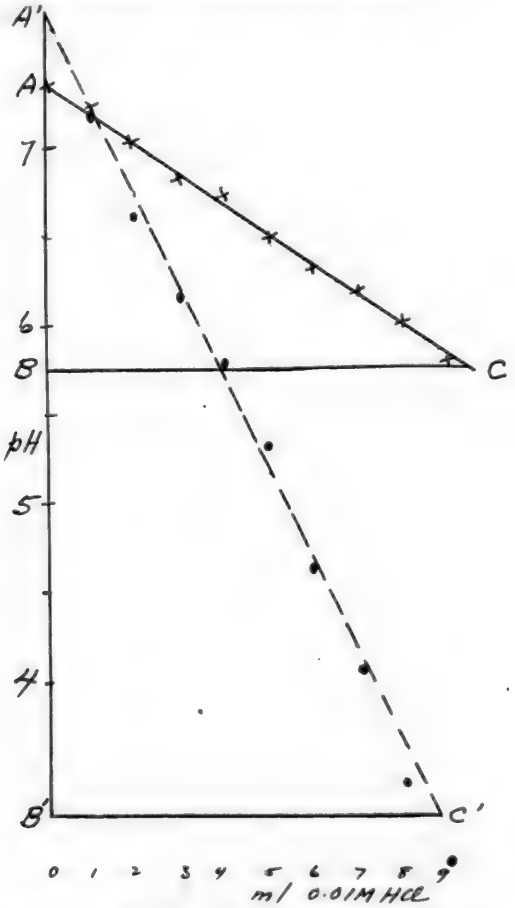

Fig. 1.—Relation between pH and volume of HCl added to saliva.

EXPERIMENTAL

(A) The buffering capacity of saliva was measured by taking 5 ml. of fresh stimulated saliva and determining its initial pH by means of a Beckmann pH meter (glass electrode). Then 1 ml. portions of 0.01M HCl were added to the saliva and the pH determined after each addition until 9 ml. had been added. The pH was then plotted against ml. of acid added and the slope of the curve thus obtained was measured. Examples of two curves near opposite ends of the series are shown in Fig. 1.

The slopes of the curves are obtained by dividing the change in pH *(AB)* by the ml. of acid added *(BC)*, e.g., in the curve labelled H.G.L. we obtain a value of 0.167. In the second example, the curve labelled J.G.H., A^1B^1 is divided by B^1C^1 to give the value 0.514.

It is interesting to note that the values for the buffering capacity so obtained show very little variation between centrifuged and uncentrifuged fresh samples of the same saliva (0.278, 0.278) and between the values for fresh saliva and saliva stored in the refrigerator for two days (0.224, 0.231).

(B) The viscosity was determined on centrifuged samples of saliva by means of a stop watch and Ostwald-Fenske viscosity pipette. Specific gravities were determined by dropping saliva into solutions of benzene and toluene of known specific gravity.[7]

(C) The salivary lactobacillus counts are in most cases the average of three successive days and were made by Miss Berry of the Dental Faculty of this University.

(D) The dental examinations were made under the direction of Dr. W. McIntosh of the same faculty.

The results are shown in Table I. Series A was determined in October and Series B in December.

TABLE I

BUFFERING CAPACITY, LACTOBACILLUS COUNT AND VISCOSITY OF SALIVA

STUDENTS INITIALS	BUFFERING CAPACITY						SALIVARY LACTOBACILLI 1000'S	RANK	ACTIVE CARIES	D.M.F.	VISC. SEC.
	SERIES A	RANK	INIT. pH.	SERIES B	RANK	INIT. pH					
J. C. B.	0.148	1	7.58	0.193	1	7.28	105	59	3	30	57.2
H. G. L.	0.167	2	7.34	0.198	2	7.38	9	21	1	14	62.4
A. G. R.	.0184	3	7.52	0.264	23	7.30	27	35	0	11	66.6
F. J. P.	0.195	4	7.45	0.310	36	7.25	60	52	0	36	58.6
J. G. C.	0.199	5	7.55	0.228	8	7.42	30	37	0	15	60.0
N. P. T.	0.201	6	7.28	0.352	46	7.35	150	62	2	20	57.0
G. E. B.	0.208	7	7.75	0.207	3	7.20	0.9	14	2	16	57.8
B. K.	0.213	8	7.60	0.243	14	7.20	12	24	0	17	58.6
H. L. G.	0.215	9	7.40				1.3	15	0	11	
A. S. U.	0.215	10	7.50	0.292	31	7.45	30	40	2	12	59.5
A. E. S.	0.216	11	7.50	0.250	17	7.40	4.5	18	4	19	55.0
L. W.	0.220	12	8.05	0.267	24	7.72	8	20	1	6	58.2
R. H. T.	0.224	13	7.30	0.350	45	7.13	158	63	6	24	58.0
R. H. F.	0.232	14	7.86	0.278	28	7.63	0	8	2	9	56.6
L. R. P.	0.235	15	7.42	0.216	5	7.03	375	70	2	11	59.5
E. A. C.	0.244	16	7.52	0.212	4	7.12	0	1	0	21	57.9
E. G. B.	0.247	17	7.75	0.259	21	7.22	12	23	2	36	53.8
D. L. A.	0.256	18	7.10	0.222	6	7.18	106	69	5	27	59.1
D. M. P.	0.260	19	7.43	0.395	58	6.96	30	41	3	33	59.3
E. J. K.	0.262	20	7.38	0.250	16	7.09	15	28	0	13	55.9
A. J. H.	0.272	21	7.90	0.228	9	7.70	260	67	0	13	61.2
G. R. G.	0.278	22	7.39	0.229	10	7.61	0	6	1	26	63.6
D. E. G.	0.279	23	8.0	0.252	18	7.80	27	33	1	17	58.4
W. G. R.	0.281	24	7.43	0.242	12	7.16	30	39	1	21	62.6
R. J. V.	0.281	25	7.78	0.227	7	7.50	90	56	2	12	58.6
B. S.	0.282	26	7.40	0.362	50	7.80	11	25	2	19	66.4
J. C.	0.284	27	7.65	0.398	59	7.80	87	55	0	12	58.1
P. E. W.	0.286	28	7.58	0.348	44	7.30	0	2	0	3	58.0
F. H. C.	0.288	29	7.50	0.252	19	7.70	0	3	0	7	58.1
R. A. M.	0.289	30	7.70				98	58	0	19	
W. A. D.	0.295	31	7.38	0.331	40	7.63	33	44	11	22	59.3
J. C. D.	0.296	32	8.01	0.290	30	7.32	45	49	4	13	
H. S. W.	0.297	33	7.30				36	46	9	43	

TABLE I (CONT'D)

STUDENTS INITIALS	BUFFERING CAPACITY						SALIVARY LACTOBACILLI 1000'S	RANK	ACTIVE CARIES	D.M.F.	VISC. SEC.
	SERIES A	RANK	INIT. pH.	SERIES B	RANK	INIT. pH					
R. S. K.	0.302	34	7.62	0.257	20	7.20	16	29	2	17	60.2
G. C. R.	0.302	35	7.65	0.299	34	7.22	9.5	22	8	44	68.0
J. E. S.	0.305	36	7.12	0.273	26	7.12	25	32	2	35	56.8
W. G. H.	0.306	37	7.58	0.279	29	7.40	13	26	5	24	65.8
J. R. W.	0.306	38	7.25	0.300	35	6.97	180	65	2	38	56.8
A. E. W.	0.308	39	7.45				14	27	1	23	
R. G. M.	0.310	40	7.62	0.317	38	7.83	0	7	1	6	58.4
T. V.	0.311	41	8.04	0.292	32	7.63					58.8
H. J. C.	0.312	42	6.90	0.355	47	7.22	175	64	2	20	
W. Y.	0.330	43	7.52	0.337	41	7.40	390	71	4	30	59.0
R. E. C.	0.333	44	7.70	0.324	39	7.20	28	34	2	32	58.2
N. P. D.	0.338	45	7.55	0.471	63	7.10	30	38	0	21	68.2
M. C. C.	0.340	46	7.84	0.277	27	7.15	47	50	3	37	60.4
M. N. B.	0.344	47	8.00	0.378	53	7.38	320	69	3	24	60.4
J. J. M.	0.353	48	7.15	0.356	48	7.10	116	61	1	13	59.6
A. A. W.	0.360	49	7.59	0.310	37	7.50	21	30	4	18	62.2
B. W. P.	0.368	50	7.50	0.368	51	7.20	82	54	0	19	62.6
G. F. M.	0.371	51	7.85	0.378	52	7.70	0.4	12	0	12	67.8
H. B. S.	0.375	52	7.08	0.487	65	6.70	230	66	3	20	70.9
J. E. H.	0.384	53	7.30	0.297	33	6.88	75	53	7	34	56.4
H. A. F.	0.410	54	7.98	0.358	49	7.6	460	72	9	29	63.4
P. K. T.	0.436	55	7.75	0.477	64	7.0	28	36	5	25	68.5
E. P. F.	0.452	56	7.58	0.393	57	7.5	0.07	11	14	26	56.6
C. M. W.	0.457	57	7.75				43	48	0	21	
J. P. M.	0.458	58	7.30	0.388	56	7.08	2	16	0	17	57.9
E. L. P.	0.500	59	7.35	0.379	54	7.10	0.05	9	0	25	61.0
J. G. H.	0.514	60	7.90	0.415	60	7.40	0	4	0	23	63.9
E. J. S.	0.531	61	7.52	0.380	55	7.43	33	45	3	34	69.2
W. R. T.	0.532	62	7.10	0.267	25	7.40	660	73	4	24	
P. M. B.				0.339	42	7.53	7	19	9	31	58.2
M. C. B.				0.260	22	7.34	0.5	13	0	11	64.4
L. V. C.				0.250	15	6.81	0	5	0	19	57.6
J. A. L.				0.437	61	7.30	22	47	0	35	62.2
J. D. G.				0.454	62	7.70	3	17	1	19	63.5
P. O. H.				0.242	13	7.53	25	31	2	25	57.8
E. W. L.				0.235	11	7.27	0.05	10	3	20	
A. R. W.				0.342	43	7.0	90	57	0	22	62.9
W. G. E.							30	43	19	4	
M. R. S.							30	42	3	37	
D. A. M.							60	51	8	49	
J. L. Y.							237	67	0	35	

CONCLUSIONS

1. The variation in buffering capacity of individual salivas in Series A and Series B is more than would be expected if there is any marked constancy in this value for a particular individual. However, there is strong evidence of positive rank correlation between the two series and high values tend to stay high and low values tend to stay low.

2. There seems to be no evidence of positive rank correlation between the buffering capacity and the salivary lactobacillus counts. In all cases the salivary lactobacillus counts were made at the same time as the saliva was taken for analysis.

3. There seems to be no significant variation in the viscosities of the saliva and with one or two exceptions they approximate 58.5 seconds. There was no

evidence of positive rank correlation between viscosity times and salivary lactobacillus counts. The densities of the saliva were remarkably constant and showed so little variation that it was considered inadvisable to calculate the viscosity on the basis of these variations which might easily be due to experimental error.

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GEOGRAPHIC VARIATIONS OF DENTAL CARIES IN OREGON

II. Dental Caries Among Institutionalized Children and the Possible Influence of Certain Ecological Factors on Its Incidence

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THE incidence of dental decay among school children has been reported in numerous articles. From these investigations it has been pointed out that the attack rates of the disease vary considerably among children living under various conditions and in different geographic areas of the country. In an effort to explain this variability of the incidence of dental decay, the influence of environmental and other factors has been analyzed and discussed.

As a part of a study which aimed to find the dental caries attack rates among school children of different geographic regions of the State of Oregon, during the winter of 1948, a dental examination was given to children living in an institution known as the Children's Farm Home of Oregon. The data reported in this paper will present and discuss the incidence of dental decay among them and some of the factors which might have influenced it.

MATERIAL AND PROCEDURE

The subjects of this study were all the children between the ages 6 to 16 years residing at the Children's Farm Home. The dental examination was conducted by means of mouth mirror and explorers in a trailer. The procedure employed for the examination and the criteria of what were considered carious lesions have been reported.[1] All examinations were made by one of us (D. M. H.); therefore, variation in caries diagnosis was avoided.

Observations were made only for permanent teeth exclusive of third molars. The dental caries experience was measured by counting the number of filled teeth, missing teeth because of caries, teeth indicated for extraction, and carious teeth indicated for filling. A tooth both carious and filled was counted only once in the total. The symbol D.M.F. was used to express the past and present dental caries experience in terms of teeth.

For a better understanding of the pattern of living of the subjects, it seems pertinent to give some information about the institution. The Children's Farm Home of Oregon, which is located three miles north of the city of Corvallis, was established in 1923 and it is sponsored by the Women's Christian Temperance Union. The Home cares for dependent children from

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This investigation was made possible through appropriations by the Oregon State Legislature in 1945 and 1947 for research under the direction of the Department of Foods and Nutrition, School of Home Economics. The Extension Women's Council of the State of Oregon was instrumental in obtaining the fund. The study was also a part of the Western Regional Research Project on the Nutritional Status of Population Groups in Selected Areas of Oregon with the cooperation of the Oregon Agricultural Experiment Station. It was financed in part from funds appropriated under The Research and Marketing Act of 1946. The cooperation and assistance of the Bureau of Human Nutrition and Home Economics, U.S. Department of Agriculture, and the U.S. Public Health Service is acknowledged.

the State of Oregon only, and it is supported by state and county funds, contributions, and the Oregon W.C.T.U. The ages of the children residing there are between 5 and 18 years, but preference for admittance is given to the younger age groups. The length of time each child spends in residence varies considerably depending upon the re-establishment of normal living conditions in his family. The capacity of the Home is about 160 children who live in eight cottages. Education is provided for all children either at the institution or at the near-by city of Corvallis. A farm of approximately 200 acres which is adjacent to the Home supplies a great amount of all the foods consumed there. The same amount and kind of foodstuffs are provided for all the children, but the preparation of the food varies depending upon the cook and the housemother in each cottage. Every week the older children receive some pocket money provided by the Home which is spent either for small personal items needed or for delicacies when visiting the near-by towns. During the summer months the children stay at the Home except on rare occasions when some of them visit with relatives and friends for a few days. Medical and dental services are provided regularly, and a registered nurse is in residence. The Home also provides toothbrushes and tooth paste to all the children at regular intervals.

FINDINGS

The total number of children examined and their distribution by age and sex are given in Table I. Altogether 163 children, between the ages 6 to 16 years inclusive, were included in the examination. The number of boys was slightly higher than that of girls.

Table II gives a picture of the period the subjects had spent in residence at the institution up to the time of the examination. This is expressed in terms of median and range by age groups, because of the wide variability in length of residence observed among the children. On this basis, the age group 6 to 7 years old had a median of 1 year and 3 months' time of residence at the institution. As it could be expected, the length of residence increased as the children advanced in chronological age and reached a median of 4 years and 6 months at the age group 14 to 16 years old.

TABLE I

NUMBER OF SUBJECTS AT THE CHILDREN'S FARM HOME GIVEN DENTAL EXAMINATION, BY SPECIFIED AGE AND SEX GROUPS

	AGE IN YEARS (LAST BIRTHDAY)											
SUBJECTS	6	7	8	9	10	11	12	13	14	15	16	ALL AGES
No. of boys	2	3	4	10	7	12	14	14	12	10	4	92
No. of girls	2	7	6	2	5	9	7	10	13	4	6	71
Both sexes	4	10	10	12	12	21	21	24	25	14	10	163

The data presented in Table III provide information per child by specified age groups on the rates for each one of the four items comprising the dental caries experience, as well as on the rates for the D.M.F. teeth. As it can be seen, the caries experience of the subjects increases simultaneously with their chronological age. While the 6-year-old child was free of dental caries, at the age of 16 years his dental caries experience had increased to an average of 8.5

TABLE II

LENGTH OF RESIDENCE TIME OF THE SUBJECTS AT THE INSTITUTION IN TERMS OF MEDIAN AND RANGE BY AGE GROUPS

AGE GROUPS	NUMBER OF SUBJECTS	MEDIAN		RANGE			
		YRS.	MOS.	YRS.	MOS.	YRS.	MOS.
6 to 7	14	1	3	0	1 to	2	8
8 to 10	34	1	7	0	3 to	4	8
11 to 13	66	2	5	0	2 to	9	1
14 to 16	49	4	6	0	6 to	11	6

TABLE III

SUMMARY OF DENTAL CARIES EXPERIENCE OF 163 SUBJECTS RESIDING AT THE CHILDREN'S FARM HOME. RATES ARE EXPRESSED PER CHILD BY SPECIFIED AGE GROUPS

PERMANENT TEETH	AGE IN YEARS (LAST BIRTHDAY)											ALL AGES
	6	7	8	9	10	11	12	13	14	15	16	
Filled	0.0	0.0	0.1	0.7	0.8	1.3	1.4	2.6	3.5	2.6	4.8	1.9
Missing	0.0	0.0	0.1	0.0	0.4	0.1	0.0	0.2	0.4	0.4	1.0	0.2
Extraction indicated	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.8	0.2	0.1
Filling indicated	0.0	0.2	1.2	1.4	2.3	2.7	2.1	3.0	4.5	5.4	3.7	2.8
D.M.F. teeth	0.0	0.2	1.3	2.0	3.3	3.7	3.4	5.0	7.2	8.4	8.5	4.5

D.M.F. teeth. In general, the average number of missing teeth and teeth requiring extraction because of extensive caries was found to be negligible. On the other hand, the average number of filled teeth per child was 1.9, while the average number of carious teeth requiring filling was 2.8, a difference of almost one tooth. This disparity between the number of filled teeth and teeth requiring filling reflects the amount of dental care received by the children at the institution. Of all the children examined thirty of them were found to be caries-free. The majority of these were among the younger age groups.

From previous investigations on the incidence of dental caries among selected groups of people in the State of Oregon,[1, 2, 3] it had been pointed out that the incidence of the disease among them was high as compared with the same age groups examined in other parts of the country. In order to be able to compare the findings of the present study with a previous one conducted in four counties of the state,[1] Table IV was prepared. It is evident from this table that the D.M.F. teeth rates of the children residing at the Children's Farm Home were considerably lower than the rates of school children examined in four counties of the state; the greatest difference was observed between the children examined in Clatsop county, and the least in those of Klamath county. Comparison of the D.M.F. teeth rate of all ages between the Children's Farm Home and each one of the four counties showed that the differences are statistically signficant with the exception of Klamath county. However, it should be remembered that the dental examination in the four counties of the state was conducted only among those children who were born and reared in their respective counties.

DISCUSSION

In an effort to explain the wide differences in the dental caries attack rates noted between the school children residing at the Farm Home and those

TABLE IV

COMPARISON OF AVERAGE NUMBER OF D.M.F. TEETH PER PERSON BY SPECIFIED AGE GROUPS OF SUBJECTS RESIDING AT THE CHILDREN'S FARM HOME AND IN FOUR COUNTIES OF THE STATE OF OREGON

AGE IN YEARS (LAST BIRTHDAY)	LOCALITY				
	CHILDREN'S FARM HOME	CLATSOP COUNTY	COOS COUNTY	DESCHUTES COUNTY	KLAMATH COUNTY
14	7.2	13.3	11.1	9.6	7.5
15	8.4	15.1	13.6	12.0	9.6
16	8.5	15.3	13.9	12.3	10.0
All ages	7.8	14.4	12.7	11.3	9.0

examined in four counties of the state, a twofold approach was undertaken. First, the fluorine content of the water supply used at the institution was determined and second, the over-all diet pattern of the subjects while in residence there was investigated.

The source of the water supply at the institution is a well approximately 65 feet deep. It has been in continuous use since some years previous to the admittance of the subject with the longest period of residence included in the present study. The fluorine content of the water was determined (C. A. S.) and it was found to be 0.3 parts per million. This small amount of fluorine in the water is certainly not the factor which can be held responsible for the low incidence of dental caries observed among the children of the institution.

The diet pattern of the subject population is in the main demonstrated in the following list which represents the typical weekly allotments of basic foods for each one of the eight cottages serving as living quarters of the children. The complement of each cottage averages twenty children and three adult personnel.

WEEKLY ALLOTMENTS OF BASIC FOODS PER COTTAGE

Eggs—10 doz.
Butter—7 lbs.
Butter substitute—10 lbs.
Cheese (American)—5 lbs.
Meat—30 to 45 lbs., according to kind
Fish—15 lbs. (twice a month, served in place of meat)
Bacon—5 lbs.
Milk—56 gals. (whole milk for drinking, top milk for cereals, etc.)
Cream—as required for ice creams, etc.
Oranges—4 to 5 doz. or the equivalent in citrus juices
Tomatoes—3 gal. canned, or all they can consume in season
Assorted canned vegetables—7 gal. All they can use in season
Assorted canned fruits—9 gal. All they can use in season
Leafy and raw vegetables—All they can consume
Potatoes—100 lbs.
Cereals—ready prepared once a week, cooked cereals other days
Bread—All they can consume

A variety of different other foods is also included in the weekly allotments. In addition, every child receives two vitamin pills daily, each one containing 800 International Units of vitamin D and 3,000 International Units of vitamin A. The use of sugar as such is unrestricted. Desserts, such as pie, cake, and pudding, are served once or twice daily. Candy bars, carbonated beverages, and sweets in general are not used often, as the sale of these items is not allowed on the grounds of the institution.

The same diet pattern always had been followed by the authorities of the institution, as great amounts of the foods consumed there are produced at the farm owned by it. The consumption of sugar, butter, and butter substitutes had been curtailed to a certain degree during the war years.

The topic regarding the influence the type of diet exerts on the incidence of dental decay has been debated for a number of years. The two main schools of thought on this subject are represented by the Iowa and Michigan groups. The former maintains that an arrest of caries is observed in persons of normal physiological state who consume a well-balanced diet, while the latter asserts that caries activity is related to excessive consumption of carbohydrates in general and refined sugars in particular, and not to the nutritional adequacy of the diet. However, while studies by a number of investigators have indicated that the restriction of sugar, either natural or refined, can effectively control dental caries, there is no conclusive evidence as yet that an optimal diet alone will influence its occurrence.[4]

From the point of view of our present knowledge on nutrition, it is generally agreed that the inclusion in our daily diet, and in specified amounts, of foods which have been classified as basic in addition to other foodstuffs of choice is a guarantee for optimum diet. From the description of the overall diet pattern presented above, it can be stated, reasonably, that the daily food intake of the subjects while in residence at the institution meets the Recommended Dietary Allowances proposed for a normal diet,[5] with the exception of citrus fruits and juices. However, in a recent survey of the ascorbic acid in plasma of a number of children residing at the institution, it was reported[6] that the mean plasma value of the entire group tested was found to be near the level which is considered to be adequate. Apparently this came about by the inclusion of foods, other than citrus fruits, which are good sources of vitamin C in the daily diet of the children.

Although there was no deliberate restriction in the consumption of refined sugars and concentrated sweets among the subjects, it seems logical to assume that there was no excessive use of them, as the case may well be in a typical American community, because the sale of candy bars, carbonated beverages, and sweets generally is not allowed on the grounds of the institution.

Moreover, the sheltered life of the subjects as well as the adequacy of their diets automatically may have served to decrease further the consumption of refined sugars and sweets in general. A similar observation was made in another study.[7]

From the foregoing prolegomena, we are inclined to conclude that the low incidence of caries observed among the institutionalized children as compared with those of the same age groups examined in four counties of the state, all other factors influencing the caries attack rates being equal, was brought about by a combination of both factors, that is, the adequacy of the diet and the restriction in the intake of refined sugars and sweets, in general.

The data obtained from this study seem to be in agreement with the work of other investigators[8-11] who observed a decrease in caries activity when an

optimum diet was provided in combination with a reduction of the excessive intake of refined sugars and concentrated sweets.

SUMMARY

A dental examination by means of mouth mirror and explorers was conducted among 163 children between the ages 6 to 16 years, inclusive, residing in an institution known as the Children's Farm Home of Oregon.

Their dental caries attack rates were found to be considerably lower in comparison with those of school children of the same age groups examined in four counties of the State of Oregon.

The fluorine content of the water supply used at the institution was determined and found to be 0.3 parts per million.

An investigation of the over-all food intake of the children while in residence at the institution revealed that their diets were nutritionally adequate. Moreover, certain reasons seem to indicate that the use of refined sugars and concentrated sweets was not "excessive."

A comparatively low incidence of caries was observed among the subjects of the study and, since these subjects had an optimum diet and a restriction in the intake of refined sugars and sweets, generally, it may have been due to either or both of the factors in combination. However, the authors believe it was brought about by a combination of both factors.

Appreciation is expressed to the members of the Dental Advisory Committee, Dr. Harold J. Noyes, Dean, University of Oregon Dental School, Dr. O. T. Wherry, member of the Oregon State Board of Health, and Dr. E. R. Abbett, representative of the Oregon State Dental Association, for their helpful suggestions and criticisms in the planning of this study and the interpretation of the results. Appreciation is also expressed to Dr. Margaret L. Fincke, Head of the Department of Foods and Nutrition at Oregon State College, for numerous consultations.

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DENTAL EFFECTS OF EXPOSURE TO FLUORIDE-BEARING DAKOTA SANDSTONE WATERS AT VARIOUS AGES AND FOR VARIOUS LENGTHS OF TIME

II. Patterns of Dental Caries Inhibition as Related to Exposure Span, to Elapsed Time Since Exposure, and to Periods of Calcification and Eruption

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THE first report in this series described the examination of 2,140 white elementary and high school children of Mitchell, South Dakota, a city of 11,000 people which accidentally fluorinated its water supply for a period of 18 months subsequent to August of 1935. Of 873 children with continuous residence, 339 between the ages of 11 and 15 years had been exposed to a mean level of about 1.15 p.p.m. of fluorides at some time prior to the eruption of their first permanent molars. No caries-inhibitory effect could be demonstrated in these children at the time of examination, about ten years after Mitchell reverted to a low-fluoride water, though about 8 per cent of them exhibited mild or very mild dental fluorosis.[1]

This report deals with several smaller groups of migrant children who, during a part of their lives before moving to Mitchell, had lived at some point in the surrounding South Dakota artesian basin and had used a Dakota Sandstone water with a fluoride concentration of 1.0 p.p.m. or more through some definite age span of at least one year's duration.

Fig. 1 shows the geographical distribution of the fluoride communities in which these children had lived. Each dot on the map represents the place of residence of one child. It will be seen that they were drawn predominately from the southern part of the South Dakota artesian basin. Most of them had lived within 50 air miles of Mitchell. This radius is indicated in Fig. 1 by a circle.

In all, 297 such migrant children were discovered. Their ages ranged from 5 to 18 years. Of these, 133 had received their fluoride exposure at some time before the age of six years, that is, before the eruption of their permanent first incisors and first molars; 152 had been exposed both before and after the age of six years; and 12 had been exposed only after the permanent first incisors and first molars had fully erupted. Since the age groups were small, data for these children were grouped in increments of three years beginning with a group aged 5 to 7 years. When so grouped the data plotted smoothly up through the age range of 14 to 16 years. These grouped data, together with that for 872 continuous-residence children similarly grouped, are summarized in Table I. For convenience, continuous-residence children are

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TABLE 1

STATUS OF THE PERMANENT TEETH, BY THREE-YEAR AGE GROUPS, OF MITCHELL, SOUTH DAKOTA CHILDREN WITH VARYING FLUORIDE EXPOSURE (1947-1948)

AGE GROUP	NUMBER OF CHILDREN	MEAN NUMBER PERMANENT TEETH	NUMBER OF TEETH				D.M.F. RATE	PER CENT WITH	
			D	M	F	TOTAL DMF		D.M.F. TEETH	FLUOROSIS
Group A (Continuous-Residence Children)									
5 to 7	217	5.4	34	0	22	56	25.8	15.0	0.0
8 to 10	208	14.5	115	8	269	392	188.5	67.8	1.4
11 to 13	215	24.7	279	61	531	871	405.1	87.9	8.4
14 to 16	171	27.9	205	41	877	1123	656.7	90.1	5.8
17 to 19	61	28.3	74	31	357	462	757.4	95.1	1.6
	872		707	141	2056	2904		65.8	3.7
Group B (Fluoride Exposure Prior to Age of 6 Years)									
5 to 7	25	5.4	2	0	5	7	28.0	12.0	0.0
8 to 10	32	14.5	11	0	18	29	90.6	40.6	9.4
11 to 13	37	23.5	22	8	88	118	318.9	86 5	24.3
14 to 16	33	27.6	51	12	142	205	621.2	84.8	36.4
	127		86	20	253	359		59.8	18.9
Group C (Fluoride Exposure Both Before and After Age of 6 Years)									
8 to 10	18	14.6	6	1	17	24	133.3	50.0	16.7
11 to 13	43	24.2	18	4	68	90	209.3	60.5	46.5
14 to 16	62	27.8	35	11	96	142	229.0	62.9	51.6
17 to 19	29	28.1	15	13	99	127	437.9	79.3	58.6
	152		74	29	280	383		64.7	47.4

designated as Group A, children with fluoride exposure wholly before eruption of the permanent teeth as Group B, and children with exposure before, during, and after eruption of the permanent teeth as Group C.

It is unfortunate that so few cases of purely posteruptive fluoride exposure occurred. The mean age of the 12 children so classified is 13.83 years; their D.M.F. rate is 191.7. Seven are caries-free. At this age a D.M.F. rate on the order of 530 would be expected in low-fluoride South Dakota children. The 12 have a mean of 5.33 years' exposure to a mean level of 2.49 p.p.m. of fluorides. For whatever they are worth the data concerning these children are quite consistent with those reported by Klein for larger groups of children with posteruptive fluoride exposures.[2-5]

Data concerning the fluoride exposures of children in Groups A, B, and C are presented in Table II, with the presumption that exposure to the current 0.2 p.p.m. of fluorides in Mitchell's water has had negligible effect. It will be seen that Group B children have had a rather uniform exposure span of about three and one-half years, and that their periods of exposure are progressively less recent with advancing age. By contrast Group C children have had a progressively longer exposure time as their ages increase, and hence a progressively shorter period of residence in Mitchell.

Their D.M.F. rates as plotted in Fig. 2 should be interpreted in the light of these facts. The rates for continuous-residence children present a smooth and quite typical parabolic curve; as previously demonstrated,[1] the short

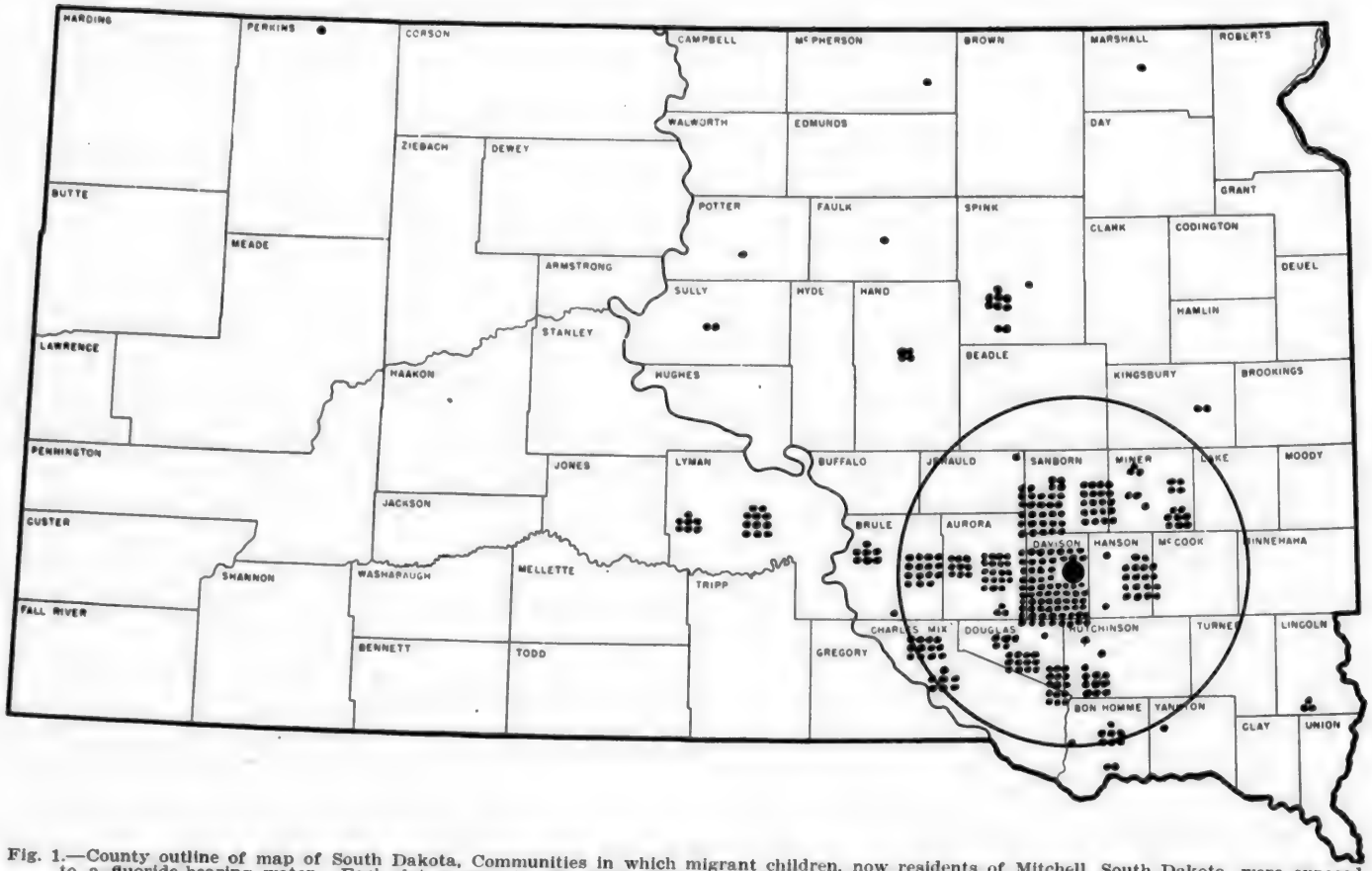

Fig. 1.—County outline of map of South Dakota, Communities in which migrant children, now residents of Mitchell, South Dakota, were exposed to a fluoride-bearing water. Each dot represents one child. The circle encloses the area within a radius of 50 miles of Mitchell.

fluoride exposure of older children in this group seems to have produced no caries-inhibitory effect which can be demonstrated after the lapse of 10 years. Caries rates for Group C children remain more or less stationary in the vicinity of two D.M.F. teeth per child.

TABLE II

MEAN FLUORIDE EXPOSURES OF GROUP A, GROUP B, AND GROUP C MITCHELL, SOUTH DAKOTA CHILDREN (1947-1948)

AGE GROUP	MEAN YEARS OF EXPOSURE*	MEAN FLUORIDE LEVEL, P.P.M.*
Group A Children		
5 to 7	—	—
8 to 10	—	—
11 to 13	.91	1.15
14 to 16	1.50	1.15
Group B Children		
5 to 7	3.44	2.01
8 to 10	3.69	2.21
11 to 13	3.57	2.07
14 to 16	3.48	1.93
Group C Children		
5 to 7	none	none
8 to 10	7.00	2.01
11 to 13	8.35	2.13
14 to 16	11.81	2.27

*Presuming that exposure to 0.2 p.p.m. of fluorides in Mitchell's present community water has had negligible effect.

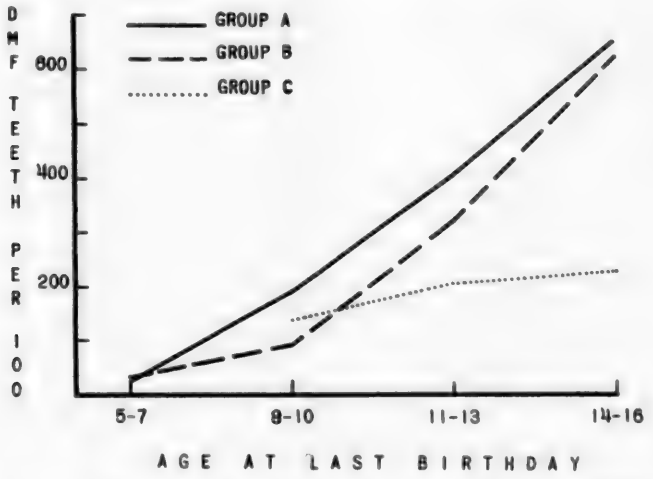

Fig. 2.—D.M.F. rates of Mitchell children with negligible fluoride exposure (Group A), with fluoride exposure terminating before the age of 6 years (Group B), and with fluoride exposure both before and after the age of 6 years (Group C).

Group B children, on the other hand, present a peculiar pattern. There would seem to be a definite caries-inhibitory effect in children aged 5 to 10 years in this group; but this effect is thereafter lost, until the prevalence of caries in Group B children aged 14 to 16 years is substantially the same as

that in Group A children with continuous residence. After the age of 10 years the incidence of new D.M.F. teeth in Group B children is actually higher than the incidence in Group A children.

This phenomenon is pictured more clearly in Fig. 3 which demonstrates the increase of new D.M.F. teeth per child between the age points plotted in Fig. 2. The data clearly suggest that, in these children at least, early fluoride exposure conferred a definite but ephemeral resistance to dental caries, and that once this resistance was lost the onslaught of caries was accelerated so that the children "caught up" with the normal prevalence of caries within the space of a few years.

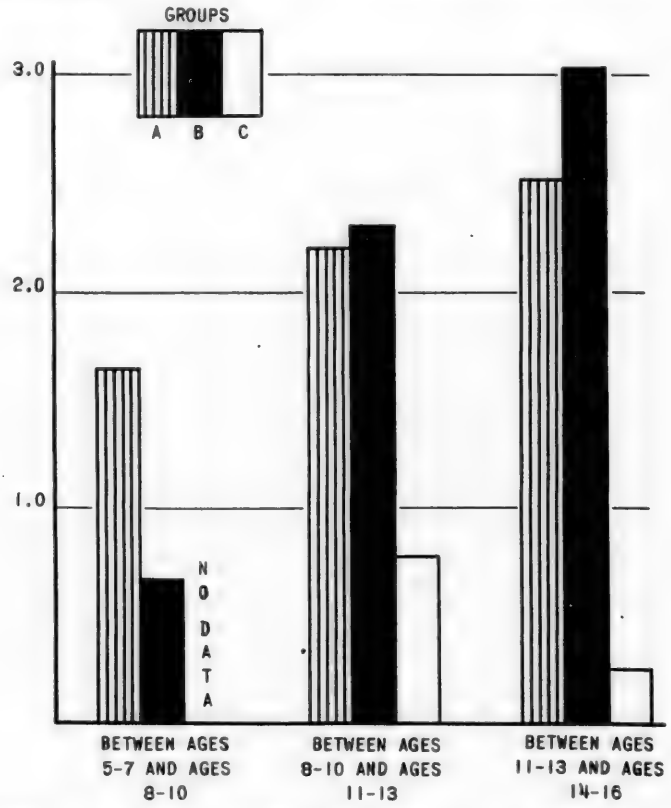

Fig. 3.—Incidence of new D.M.F. permanent teeth between the age groups plotted in Fig. 2 for three groups of Mitchell children with varying fluoride exposure.

Since these groups are small the matter of statistical significance requires careful attention. Analysis suggests that Figs. 2 and 3 may safely be taken at their face value. By the common variance approach the difference between the D.M.F. rates of children in Groups A and B is minute and unimportant at ages 5 to 7, becomes highly significant at ages 8 to 10, diminishes at ages 11 to 13, and becomes inconsequential at ages 14 to 16, where the odds are about 7 to 3 that the slight difference is due to chance.

If standard error technics are preferred the analyses may be based upon percentages of D.M.F. first molars; these teeth account for 90 per cent of all those carious in both Group A and Group B children aged 8 to 10 years. At this age point the odds are greater than 1,000 to one that the difference is not due to chance. In children aged 14 to 16 years the first molar D.M.F. rates are virtually identical and the probability is overwhelming that chance accounts for the slight difference. A detailed discussion is omitted for brevity here, since very similar data will be presented in the next segment of this report.

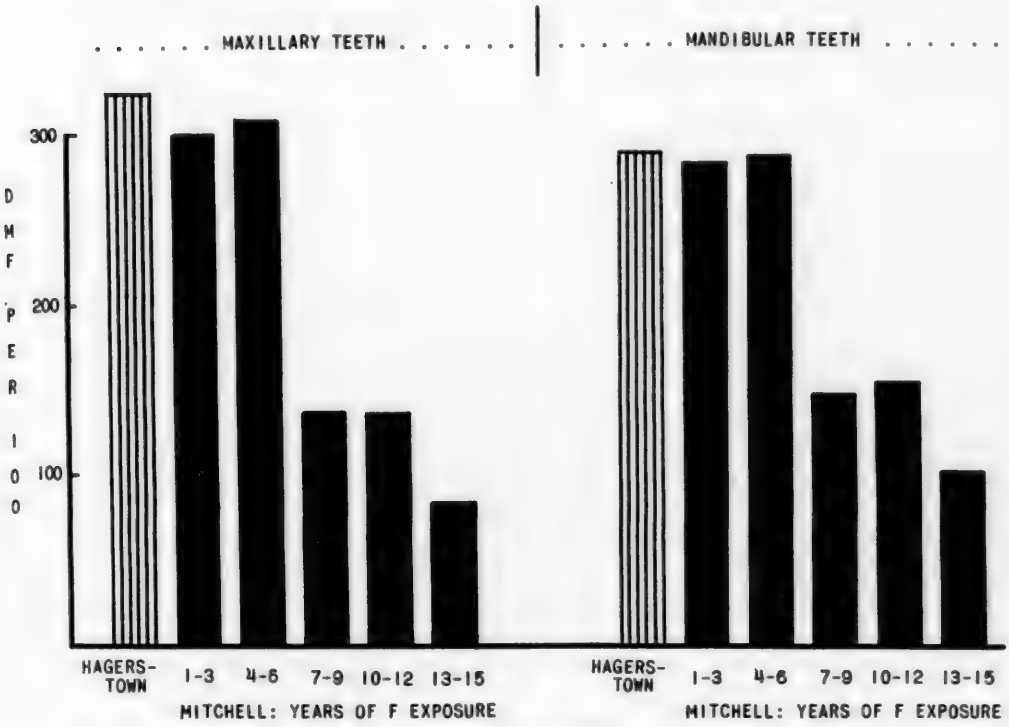

Fig. 4.—Over-all D.M.F. rates of Hagerstown children, aged 13 to 17 years, compared with D.M.F. rates in Mitchell migrant children with varying spans of fluoride exposure beginning in each case at birth.

Of these analyses, that of first molars alone is probably the more meaningful. The fluoride exposure of virtually all of these children began at birth and its mean span includes the entire period commonly accepted as the calcification time of the first molar crown. If the full caries-inhibitory effect of the fluorides follows exposure during the calcification period alone, the inhibitory effect seen in children aged 5 to 10 years in Fig. 2 should have persisted past the age of 16 years.

Among those migrant children whose fluoride exposure began at birth and continued without interruption for at least one year, there were 143 aged 13 to 17 years. Their exposure ranged from one to 17 years, to a mean fluoride

level slightly in excess of 2.0 p.p.m. Their mean age is 14.99 years; about 42 per cent show evidence of fluorosis. D.M.F. rates were calculated for children in this group with fluoride exposures of from 1 to 3, 4 to 6, 7 to 9, 10 to 12, and 13 to 15 years. For comparison D.M.F. rates of grouped Hagerstown children aged 13 to 17 years were calculated.[6] The mean age of the Hagerstown group is 14.75 years. The main difference between the groups is a disparity in sex composition; about 52 per cent of the Hagerstown group and about 61 per cent of the Mitchell group are girls. Their D.M.F. patterns are detailed in Table III and pictured in Fig. 4.

While it is obvious that extended fluoride exposure exerted a more marked caries-inhibitory effect than exposures of short duration, it is equally obvious that caries rates in these children do not decrease uniformly as exposure time is increased. In general, Mitchell migrants with fluoride exposures ranging from one to 6 years have about the same prevalence of caries as do Hagerstown children, though there is a slight and perhaps illusory advantage in the maxillary teeth. At this point the D.M.F. rates dip sharply, to level off again through exposure spans up through 12 years; whereupon another sharp dip occurs. These dips are statistically significant, and are associated with the eruption periods of the five tooth types which make up the major fraction of teeth found carious in children of this age—the maxillary first incisor and the maxillary and mandibular first and second permanent molars. Inspection of the raw data indicated that the differences between the 1 to 6- and 7 to 12-year exposure spans were probably due to a more favorable caries experience in first molars in the latter group, and that the differences between the 7 to 12- and 14 to 16-year exposure spans were probably due to a more favorable caries experience in second molars in the latter group. This suggested a new breakdown for analysis in which the fluoride exposure span could be related to the calcification and eruption periods of the specific tooth types presented by this group of children.

For this analysis the specific tooth types of these 143 children were pooled and regrouped. In one group (Group D) were placed all those teeth with

TABLE III

DENTAL STATUS OF HAGERSTOWN CHILDREN AGED 13 TO 17 YEARS, AND OF MITCHELL, SOUTH DAKOTA MIGRANT CHILDREN AGED 13 TO 17 YEARS WITH FLUORIDE EXPOSURE BEGINNING AT BIRTH

YEARS OF FLUORIDE EXPOSURE	NUMBER OF			NUMBER WITH FLUOROSIS	PER CENT D.M.F.	D.M.F. RATES IN		
	BOYS	GIRLS	TOTAL			ALL TEETH	UPPER TEETH	LOWER TEETH
Hagerstown Children								
—	1303	1408	2711	—	—	617.4	325.9	291.5
Mitchell Migrants								
1 to 3	13	11	24	5	87.5	583.3	300.0	283.3
4 to 6	9	17	26	9	96.2	596.2	307.7	288.5
7 to 9	4	15	19	11	68.4	284.2	136.8	147.4
10 to 12	20	16	36	17	75.0	291.7	136.1	155.6
13 to 15	8	24	32	15	56.3	187.5	84.4	103.1
	54	83	137	57	75.9	375.2	185.4	189.8

fluoride exposure ranging from a minimum of two-thirds of the accepted calcification period to a maximum of exposure up to the point of eruption. This exposure period presumably covered, as a minimum, the period during which caries-susceptible tooth surfaces—i.e., the occlusal surfaces and the area of the contact points, and buccal and lingual pits—were being formed. In the second group (Group E) were placed those teeth with fluoride exposure well past the period of eruption. In the case of an individual child with fluoride exposure from birth through the age of 8 years, for example, the first molars were included in Group E but the second molars were placed in Group D.

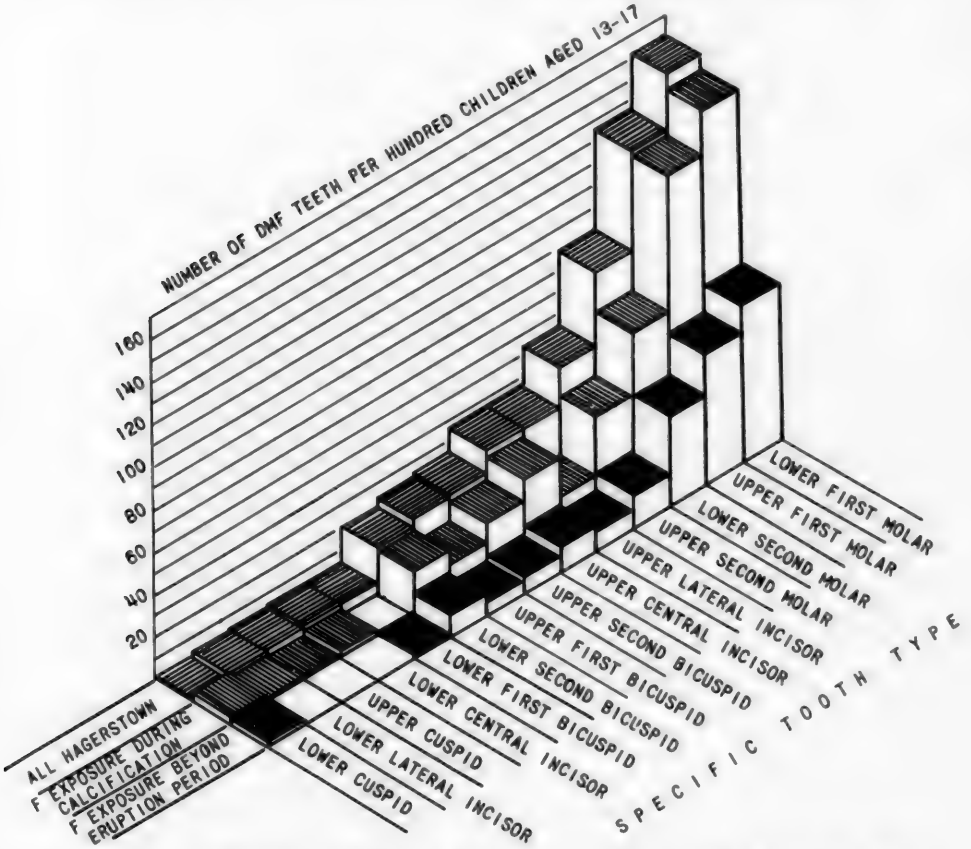

Fig. 5.—Tooth-specific D.M.F. rates of Hagerstown children, aged 13 to 17 years, compared with rates in the teeth of Mitchell migrant children aged 13 to 17 years with fluoride exposure during the major portion of the calcification period (Group D), and with fluoride exposure past the period of eruption (Group E).

The Hagerstown tooth-specific D.M.F. rates for children aged 13 to 17 years, together with those for Group D and Group E teeth and the exposure-span criteria by which they were classified, are shown in Table IV. The relative magnitude of these rates is shown in Fig. 5. Although the entire array of rates is given in Table IV and in Fig. 5, the Mitchell sample is too small for

significance in any but the six most caries-susceptible tooth types—the upper first and second incisors and the first and second molars—and the apparent plausibility of the pattern established by the less susceptible tooth types may be spurious.

However, these susceptible teeth constitute the most serious dental health problem in children of this age, and are sufficient to establish the few generalizations required for the present purpose. It will be seen that, with the exception of the upper second incisor, fluoride exposure during the major portion of the calcification time conferred a uniform but slight resistance to dental caries, but that this advantage is negligible in those teeth which have been longest at risk of caries—the first incisor and the first molars. On the contrary, teeth with a fluoride-exposure span extending well past the period when those teeth have erupted show a more favorable caries experience in every case, with a magnitude of caries inhibition very much like that seen by Dean in his studies of 12- to 14-year-old children with continuous residence in a fluoride area.[7]

TABLE IV

TOOTH-SPECIFIC D.M.F. RATES IN HAGERSTOWN CHILDREN AGED 13 TO 17 YEARS, AND IN THE TEETH OF 143 MITCHELL, SOUTH DAKOTA MIGRANT CHILDREN AGED 13 TO 17 YEARS WITH FLUORIDE EXPOSURE BEGINNING AT BIRTH AS RELATED TO PERIODS OF CALCIFICATION AND ERUPTION

TOOTH TYPE	HAGERSTOWN RATE	GROUP D*			GROUP E†		
		NUMBER CHILDREN	YEARS F EXPOSURE	D.M.F. RATE	NUMBER CHILDREN	YEARS F EXPOSURE	D.M.F. RATE
Maxillary Teeth							
Second molar	56.0	54	5 to 11	40.7	27	14 to 17	14.8
First molar	136.6	35	2 to 5	134.3	90	8 to 17	60.0
Second premolar	27.0	45	4 to 9	22.2	38	13 to 17	7.9
First premolar	24.9	57	3 to 9	14.0	54	12 to 17	5.6
Canine	5.0	71	3 to 10	1.4	38	13 to 17	--
Second incisor	38.3	41	3 to 7	12.2	74	10 to 17	8.1
First incisor	38.2	43	2 to 6	34.9	80	9 to 17	11.3
Mandibular Teeth							
Second molar	91.6	48	5 to 10	70.8	27	14 to 17	40.7
First molar	162.5	35	2 to 5	154.3	90	8 to 17	73.3
Second premolar	21.6	59	4 to 10	20.3	38	13 to 17	10.5
First premolar	5.3	34	5 to 9	--	38	13 to 17	2.6
Canine	.8	51	3 to 8	3.9	60	11 to 17	1.7
Second incisor	4.2	43	2 to 6	2.3	80	9 to 17	--
First incisor	5.3	43	2 to 6	3.1	90	8 to 17	--

*Teeth with fluoride exposure ranging from a minimum of two-thirds of the commonly accepted calcification period up to the point where eruption begins.

†Teeth with fluoride exposure from birth past the period of eruption.

Compared with the Hagerstown pattern by chi square analysis in which E = the number of D.M.F. teeth which would have appeared had the Hagerstown rates obtained, the Group D pattern is not significantly different if the upper second incisor is excluded. The Group E pattern is strikingly different; it yields a chi square total of 440.5, much greater than the value of 34.5 required to establish chance odds of one in 1,000 with 13 degrees of freedom.

The inference is plain that, in these children at least, fluoride exposure past the period of eruption and well into the time when the teeth were exposed in the mouth was required for any marked or important caries inhibition in those caries-susceptible tooth types which present the major dental problem for children of these ages.

DISCUSSION

The patterns of caries inhibition in these children are definite, clear, and significant. They suggest the following as a working hypothesis:

A. Fluorides may be incorporated into tooth enamel either during calcification or after eruption, and when so incorporated are effective in the inhibition of dental caries.
B. This inhibitory effect tends to persist so long as fluoride exposure is continued but tends slowly to be lost after fluoride exposure is discontinued; hence
C. Periodic or continuous renewal of the fluoride content of tooth enamel is required for maintenance of the maximum caries-inhibitory effect.

Proposition (B) of this hypothesis is in direct conflict with the findings of a study made at Bauxite, Arkansas, some 12 years after that community changed from a fluoride-bearing to a fluoride-free water supply.[8] In the 24 children aged 13 to 17 years found in Bauxite in 1940, about 31 per cent of first molars were found carious. This is very similar to the first molar attack rate in Group E teeth in this study (33 per cent), and dissimilar to the first molar attack rate in Group D teeth (72 per cent).

There are two distinct differences between the two studies which might explain these contradictory findings. For one, exposure at Bauxite was to a fluoride concentration of about 14 p.p.m., which might have had a more lasting effect than exposure to concentrations in the neighborhood of 2.0 p.p.m. as seen in Mitchell migrants. For another, the investigators at Bauxite recorded an inhibition of caries in a "transitional group" of children, most of whom had had no direct fluoride exposure at all. This suggests that other and unrecognized caries-inhibitory forces might have been at work in the Bauxite study children.

With this outstanding exception, virtually all of the evidence in the literature is compatible with the hypothesis as stated. The concept of a lasting inhibition of caries following fluoride exposure during the calcification period seems to have been based at least in part upon the results of animal experimentation; but Hodge and Sognnaes, reviewing this field in toto, concluded that fluorides were more effective in inhibiting rat caries when administered after tooth eruption than when given during the pre- and postnatal periods alone.[9] Particularly compelling is the manner in which these findings complement the findings of Klein in his studies of individuals with precisely the opposite exposure experience, that is, birth in a low-fluoride area and subsequent migration to a community with a fluoride-bearing water supply.[2-5]

In the area of fluorination of community water supplies it has been assumed that the benefits of fluorination, if any, would be confined to that segment of the population eight years of age or younger. This has raised the question as to whether some more specific and direct mode of fluoride administration might not be preferable to mass fluorination of community water supplies.[10] In fact, one recent reviewer makes this flat statement: "There is agreement that fluorine, and by inference, other chemicals cannot prevent caries in the erupted tooth . . . it is evident that application of fluorine or chemicals to the teeth or the ingestion of them in drinking water or in the form of tablets is not a practical method of prevention of caries."[11] On the contrary, these findings imply that fluorination may benefit individuals up through the age of 17 years, and, possibly, all individuals of the community of whatever age. If this is true the entire area of the practicability of mass fluorination must be re-examined.

None of these findings apply directly to teeth treated topically with a fluoride solution, and inferences may be fallacious if it should develop that the caries-inhibitory mechanism set up by topical treatment is essentially different from the caries-inhibitory mechanism which follows the exposure of a tooth in eruption to a fluoride-bearing drinking water. However, if these mechanisms are identical or essentially similar it would seem to follow that the caries-inhibitory effect which follows topical treatment may likewise prove ephemeral; that repetition of treatments may be required for maintenance of the maximum inhibitory effect; and that more frequent repetition may be required for the protection of highly caries-susceptible than for relatively caries-immune tooth types. These propositions can be tested by continued observations of children already treated experimentally with sodium fluoride, with particular attention to the patterns of caries inhibition in specific morphological tooth types.

This should be regarded as a preliminary report. The study was designed to demonstrate an expected caries-inhibitory effect in those continuous-residence (Group A) children who had undergone an 18-months' period of accidental fluoride exposure in 1935, 1936, and 1937. The significance of these caries-inhibitory patterns in migrant children did not emerge until the latter stages of statistical analysis were reached, and it is pure accident that these children should have presented in sufficient numbers that the patterns discussed in this report are valid from the statistical point of view. Despite statistical significance it should be emphasized that these findings are based upon comparatively small groups of children who are residents of a limited geographical area. However, since their implications are clear and important, it is hoped that the suggested hypothesis will be considered and tested by workers in other geographical areas and in other fields of study.

SUMMARY AND CONCLUSIONS

1. Mouth-mirror examinations were given 2,140 white children of Mitchell, South Dakota, a community which accidentally fluorinated its water for a

period of 18 months during 1935, 1936, and 1937. No inhibition of dental caries could be demonstrated, ten years later, in continuous-residence children who had used this water.

2. Among migrant children with fluoride exposure, those with exposure wholly prior to eruption of the first permanent molars showed a transient inhibition of dental caries in those teeth which persisted through the age of 10 years but was virtually lost by the age of 14 to 16 years. Caries-inhibitory effects persisted, undiminished in magnitude, through the age of 14 to 16 years in those children whose fluoride exposure continued past the time of eruption of the first permanent molars.

3. In migrant children 13 to 17 years of age whose fluoride exposure began at birth, tooth types with fluoride exposure only through the period of calcification presented a slight and perhaps illusory inhibition of caries. Caries reduction in teeth highly susceptible to caries was marked only when fluoride exposure was continued for some time after eruption of those teeth.

4. These findings become coherent if the following working hypothesis is essentially true:

A. Fluorides may be incorporated into tooth enamel either during the process of calcification or after eruption and when so incorporated are effective in the inhibition of dental caries.

B. This inhibitory effect tends to persist so long as fluoride exposure is continued but tends slowly to be lost after fluoride exposure is discontinued, particularly in teeth highly susceptible to caries; hence

C. Periodic or continuous renewal of the fluoride content of tooth enamel is required for maintenance of the maximum caries-inhibitory effect.

5. This working hypothesis is offered for verification, modification, or disproof by other workers in other geographical areas and in other fields of study.

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SALIVARY CITRATE AND DENTAL EROSION

Procedure for Determining Citric Acid in Saliva—Dental Erosion and Citric Acid in Saliva

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PART I

CITRIC acid has been studied in a number of tissues and body fluids, but only to a limited extent in saliva. Pucher, Sherman, and Vickery[1] found 0.04 to 1.30 mg. per cent in seven saliva specimens, but Kuyper and Mattill[2] and Leake[3] were unable to detect citric acid in saliva using methods, however, which were not sensitive to less than 2.0 mg./100 ml. Zipkin[4] reported saliva (some 180 analyses on 15 adult men) to contain 0.50 to 2.00 mg. per cent citric acid, expressed as the monohydrate. Recently, Shulman and Robinson[5] used the gravimetric method of Duysher and Holm[6] and found 0.0 to 1.55 mg. per cent citric acid in the saliva of 25 normal individuals. In general, the gravimetric procedure is not as sensitive or as specific as the colorimetric technic.[7]

Following a survey of methods for citric acid in biological fluids, the colorimetric procedure of Perlman, Lardy, and Johnson,[8] with several modifications, was found satisfactory for the analysis of 10 ml. specimens of saliva. Our experience with this method is somewhat contrary to Shulman and Robinson,[9] however, who reported negative results for a number of saliva specimens. Recently, Natelson, Lugovoy, and Pincus[10] used essentially the technic employed by us. Speck, Moulder, and Evans[11] did not modify the original Perlman, Lardy, Johnson procedure[8] and discarded it as unsatisfactory.

A critical analysis of this citrate method[8] and its application to saliva analysis will be reported prior to presenting clinical data on the relation of citric acid in saliva to dental erosion.

Current interest in salivary citrate relates to the observation that rats' dental tissues are attacked in vivo by nearly neutral citrate drinking fluids[12] thus suggesting a role of salivary citrate in human dental erosion.

Procedure for Citric Acid in Saliva.—About 15 ml. of paraffin-stimulated saliva were collected in a 35 ml. glass-stoppered brown bottle using a short-stemmed funnel. Each bottle contained 0.5 ml. of concentrated H_2SO_4. After thorough shaking, a 10.0 ml. aliquot was transferred to a test tube containing 2.0 ml. of 18N H_2SO_4 and placed in a boiling water bath for 20 minutes. The

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sample was filtered hot and washed with 5.0 ml. of hot water. Five-tenths milliliter of saturated bromine solution was then added and the sample allowed to stand for 2 hours in the refrigerator for complete precipitation of bromination products. The mixture was filtered and washed with 5.0 ml. of hot water, the filtrate and washings being received in a 50 ml. graduated centrifuge tube. Consecutively were added 0.3 ml. 18N H_2SO_4, 0.2 ml. 1M KBr solution, and 0.75 ml. saturated $KMnO_4$ solution (70 Gm./1). The mixture was allowed to stand for 15 minutes. Three per cent H_2O_2 was added just to disappearance of MnO_2, and 0.1N $KMnO_4$ was then added to destroy the excess H_2O_2.

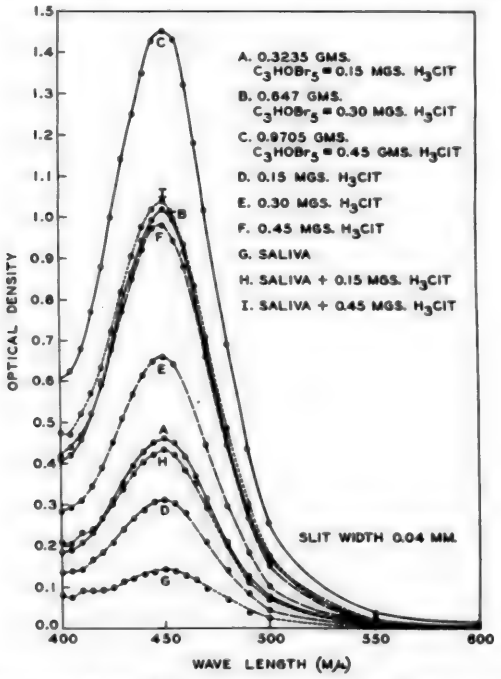

Fig. 1.—Spectral transmittance curves for various concentrations of pentabromacetone, citric acid, and citric acid in saliva using a Beckman model DU quartz spectrophotometer.

The clear and colorless solution was made up to 30 ml. with distilled water, 13 ml. of Skellysolve B were added, and the sample shaken vigorously for 15 seconds. The sample was centrifuged for a few minutes at about 1,000 r.p.m. to break the emulsion, and 10 ml. of the supernatant liquid were separated by aspiration and transferred to a colorimeter tube. Three milliliters of a 50 per cent dioxane-water mixture were added, followed by 3.0 ml. of 4 per cent sodium sulfide solution freshly prepared and filtered. The sample was thoroughly shaken for 15 seconds and centrifuged for a few minutes at 1,000 r.p.m.

The yellow color was measured in a Coleman 11A spectrophotometer at 450 mμ using a blank consisting of 10 ml. of water, carried through the same procedure as the samples. Transmittance readings were converted to mg. per cent by reference to a standard calibration curve.

Preparation of Calibration Curve.—Preparation of spectral transmittance curves for various concentrations of pentabromacetone, citric acid, and of citric acid in saliva shows the maximum density to be at 450 mμ in every case (Fig. 1).

A white crystalline pentabromacetone prepared according to the method of Dickens[13] and melting at 73.5 to 74.5° C. was dissolved in Skellysolve B and the resulting solution used to study the spectral characteristics of the yellow color developed in the procedure described (Curves *A*, *B*, and *C*, Fig. 1). Curves *D*, *E*, and *F* (Fig. 1) were obtained from standard solutions of citric acid containing 0.15, 0.30, and 0.45 mg. citric acid respectively. To obtain the calibration curve, Fig. 2, a blank and 5 citric acid standards were prepared in

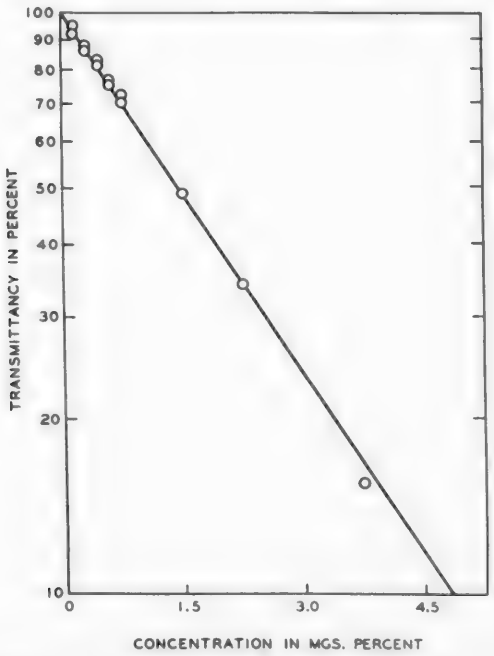

Fig. 2.—Calibration curve for citric acid in saliva using a Coleman model 11A spectrophotometer.

duplicate to contain 0.015, 0.030, 0.045, 0.060, and 0.075 mg. citric acid. A third series of standards was prepared containing 0.15, 0.225, and 0.375 mg. citric acid. Standards prepared at subsequent intervals conformed closely to the calibration curve, thus substantiating its validity.*

Evaluation of Solvents Used in Developing the Final Yellow Color.—In the production of the final yellow color, various solvents, in addition to pyridine,[1] dioxane,[14] ethylene glycol,[15] and commercial glycerol,[16] already reported, were used in an attempt to intensify the color and increase its stability.

*Our studies indicate that the calibration curve for citric acid is not a straight line throughout the entire range investigated, but, for the purposes of the study, the curve as shown is entirely satisfactory.

Solvents closely related to both pyridine and dioxane were tried, that is, piperidine (50-50), morpholine (50-50), piperazine hydrate (saturated solution), and trioxane (saturated solution). Only dioxane, pyridine, and trioxane showed any promise, and of these, dioxane-water solutions gave the greatest optical density. Both technical and redistilled grades of Skellysolve B were tried and both gave comparable results, thus obviating the necessity for purification.

When glycerol and ethylene glycol in 50-50 water mixtures were substituted for the dioxane-water mixture, they gave a color essentially stable for 60 minutes, but the optical density obtained was not as great as with dioxane-water mixtures. Various grades of dioxane gave such variable results that purification[17] was found necessary.

Relation of Sodium Sulfide Concentration to Optical Density of Yellow Color Produced.—It was found necessary to prepare the sodium sulfide solution fresh daily. A progressive decrease in the optical density of the final yellow color without any appreciable change in the density of the sodium sulfide solution was found, as the latter aged. Hence, the optical density of the sodium sulfide solution apparently cannot be used as a criterion of constancy, as reported in the literature.[10]

Attempts to prepare a satisfactory combined reagent of four per cent sodium sulfide in 50-50 dioxane-water solution were unsuccessful in that a progressive decrease in the optical density of the final yellow color was again observed as the combined reagent aged.

It was confirmed that the maximum optical density of the final yellow color is produced with a 4 per cent sodium sulfide solution. The optical density increased up to a concentration of 4 per cent sodium sulfide and then decreased.

Collection, Preservation, and Deproteinization of Saliva.—Preliminary studies showed that the citric acid concentration in saliva decreased rapidly on standing. Various bactericidal agents were studied in an attempt to stop bacterial action in the saliva immediately as collected. Among these were the following: phenylmercuric acetate, hydroxide, bromide, and salicylate; thymol; commercial formalin; and quaternary ammonium salts. None of these agents, however, was without interference in the subsequent citric acid determination; 0.5 ml. concentrated H_2SO_4 in about 15 ml. of saliva was finally adopted as the stabilizing agent, after a number of studies had demonstrated the stability of citric acid in saliva in the presence of H_2SO_4. As shown in Fig. 3, there is a striking decrease in salivary citric acid, particularly during the first hour, but when sulfuric acid is present the citric acid remains essentially constant. Curves A and B represent two different saliva specimens stabilized with H_2SO_4, and Curve C represents a specimen of saliva without H_2SO_4.

Saliva appears somewhat unique insofar as it is not easy to deproteinize with reagents commonly used for this purpose. Many of the usual deproteinizing reagents were studied including acetic acid, trichloracetic acid, tungstic acid, metaphosphoric acid, phosphotungstic acid, phosphomolybdic

acid, sulfosalicylic acid, perchloric acid, copper sulfate-sodium carbonate-sodium tungstate mixture, 95 per cent alcohol, heat, and sulfuric acid. Only the sulfuric acid proved entirely satisfactory. It was found that 2.0 ml. of 18N H_2SO_4 added to 10.0 ml. of saliva and placed in a boiling water bath 20 minutes gave a flocculent precipitate which was readily removed by filtration. During this process the filtrate usually varied in color from pink to brown, but this did not interfere in subsequent procedures.

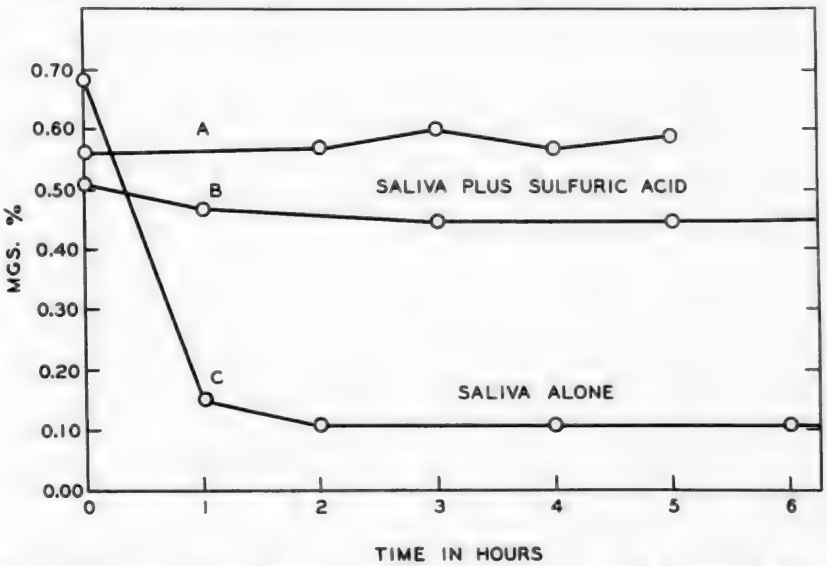

Fig. 3.—Stability of citric acid in saliva with and without sulfuric acid. Samples A, B, and C were collected from the same individual at different times.

Recovery of Citric Acid Added to Saliva.—Results of a number of recovery tests are shown in the following table:

TABLE I

RECOVERY OF CITRIC ACID* ADDED TO SALIVA

TRIAL NUMBER	CITRIC ACID PRESENT IN 10 ML. OF SALIVA (MG.)	CITRIC ACID ADDED TO SALIVA (MG.)	CITRIC ACID FOUND (MG.)	CITRIC ACID RECOVERED (MG.)	RECOVERY (PER CENT)
1	Blank	--	--	--	--
2	0.066	0.000	0.066	--	--
3	0.066	0.075	0.143	0.077	102.6
4	0.066	0.150	0.218	0.152	101.3
5	0.066	0.225	0.304	0.238	105.7
6	0.066	0.300	0.360	0.294	98.0
7	Blank	--	--	--	--
8	0.071	0.000	0.071	--	--
9	0.071	0.075	0.141	0.070	93.5
10	0.071	0.150	0.210	0.139	92.8
11	0.071	0.225	0.284	0.213	94.7
12	0.071	0.300	0.360	0.289	96.4

*Expressed as the monohydrate.

Notes on the Procedure.—

A. Five-tenths milliliter of saturated bromine solution was sufficient for bromination. Although salivas varied in the amount of bromine actually consumed, this was not related to the citric acid present.

B. A light-yellow color may appear after addition of the dilute permanganate but this was destroyed by adding a fraction of a drop of the H_2O_2.

C. Shaking of samples during extraction of pentabromacetone from water solution with Skellysolve B and later extraction from Skellysolve B with sodium sulfide and dioxane was standardized to 50 manual shakes, or about 15 seconds.

D. Purification and redistillation of commercial Skellysolve B was unnecessary.

E. Although absorption spectra were determined with a Beckman Model DU quartz spectrophotometer, the Coleman Model 11A instrument was used in routine analyses to obviate the necessity of separating the lower colored layer. Standard citric acid solutions showed a maximum density of 450 mμ with both instruments.

F. By developing the final color with 3 ml. of sodium sulfide solution and 3 ml. of dioxane-water solution instead of 5 ml. of each as originally used by Perlman, Lardy, and Johnson, an increase in sensitivity of 67 per cent was obtained.

Part II of this study is an investigation of the relation between human dental erosion and the citric acid content of saliva.

PART II

Dental Erosion.—Dental erosion has been known for almost two centuries,[18] but its etiology is still obscure. The majority of the theories which have been proposed to explain erosion have been reviewed by Thoma[19] and by Miller and Newman.[20] Erosion has also been studied by Black,[18] Darby,[21] Kirk,[22] W. D. Miller,[23, 24] Bunting,[25] Badanes,[26] Bödecker,[27] and Robinson.[28]

A dissolution of tooth substance in vivo, which resembles erosion in appearance, has been attributed to various acids in both human[29-33] and laboratory animal[34-37] observations.

As already noted, the destructive action of citrate drinking fluids on rats' molar tooth surfaces in vivo[12] suggested citrate in oral fluids localized on tooth surfaces as a cause of human erosion.

For purposes of this clinical study, erosion was defined as a loss of tooth substance at the gingival third of the buccal or labial surface, leaving a hard glossy surface. In nearly all cases, erosion was seen as a flattened or disk-shaped area on the buccal surfaces causing the normal convexity of the tooth to be lost. The degree of erosion was arbitrarily evaluated as none, mild, moderate, or severe, and designated 0, 1, 2, and 3, respectively. The "Erosion Index" is the total score divided by the number of teeth. This method of scoring is similar to that used by Restarski, Gortner, and McCay,[38] for scoring the molar teeth of rats.

This study relates to 38 individuals with an established diagnosis of erosion, and 83 individuals selected at random at this station. Among the latter group 22 had some erosion, making a total of 60 individuals with erosion and 61 with no erosion.*

Although the groups are small, it is interesting to note that among the 83 random-selected individuals, 22 (27 per cent) had some degree of erosion. Among 42 individuals, aged 27 to 39 years, 9 (21 per cent) had erosion. There

*We are indebted to Robert C. Likins, Senior Assistant Dental Surgeon (R), USPHS and to H. Berton McCauley, Senior Assistant Dental Surgeon (R), who performed the dental examinations.

were 13 erosion cases (32 per cent) among the remaining 41 individuals, aged 40 years and over.

The erosion data when studied according to prevalence in individual mouth quadrants indicate that there is probably a greater prevalence of erosion in the upper than in the lower quadrants. The mean difference between upper and lower teeth was 2.017 times its standard error, giving a probability of less than 0.05.

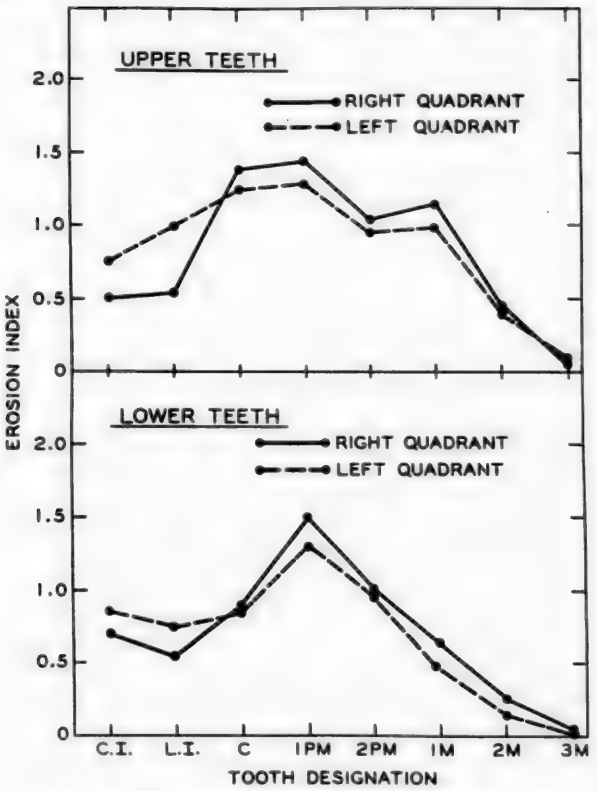

Fig. 4.—Evaluation of degree of erosion.

Analysis of the data on severity of erosion (erosion index) shows that the upper teeth are also more severely affected than the lower teeth. The mean difference between the upper and lower teeth was 2.5 times its standard error, giving a probability of less than 0.02. These data are presented graphically in Fig. 4.

The average erosion index figures not shown in Fig. 4 are as follows: upper left 0.92, upper right 0.89, and all uppers 0.90; lower left 0.73, lower right 0.78, and all lowers 0.75. The average erosion index for all teeth is 0.82. There was no significant difference in the degree of erosion in right and left quadrants of either upper or lower jaws. The first premolars were most severely affected, and the upper first molars were approximately twice as badly eroded as the lower first molars. This may be suggestive of a specific

effect of the parotid saliva, since the parotid ducts open in the area of the upper first molars.

Fig. 5 shows the relation of age to the erosion index, together with the correlation coefficient, +0.33, and the corresponding regression line, $Y = 0.018X$. The probability of occurrence by chance of this correlation coefficient is less than 0.01 indicating that the degree of erosion increases with age. It is interesting to note in this case that the regression of age on erosion index has a zero intercept.

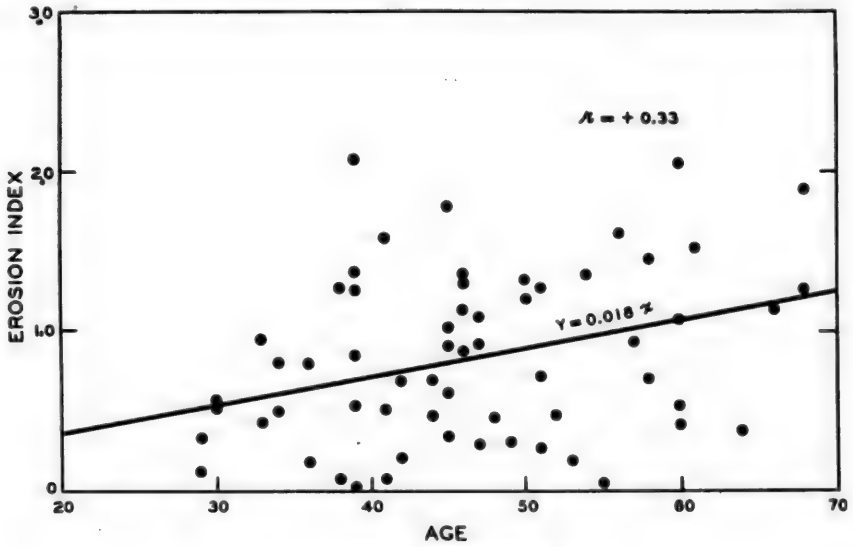

Fig. 5.—Relation of age to erosion index.

Results of Citric Acid Analyses of Saliva.—Fifteen milliliter specimens of paraffin-stimulated saliva were collected at 11 A.M. and 3 P.M. in small brown glass-stoppered bottles containing 0.5 ml. of concentrated sulfuric acid.* Citric acid was determined in both specimens according to the procedure already outlined, and the two results were averaged to obtain a representative citric-acid figure.

A summary of these analytical data is presented in Table II.

TABLE II

RELATION OF CITRIC ACID CONTENT OF SALIVA TO AGE

	NORMAL CASES				EROSION CASES			
AGE GROUP IN YEARS	NO. CASES	MEAN AGE	RANGE IN MG. % CITRIC ACID	MEAN* CITRIC ACID	NO. CASES	MEAN AGE	RANGE IN MG. % CITRIC ACID	MEAN* CITRIC ACID
27 to 39	33	32.8	0.29–1.82	0.86±0.06	17	35.5	0.48–1.67	1.04±0.09
40 to 49	15	43.8	0.45–2.09	0.99±0.11	22	44.9	0.20–1.97	1.03±0.09
> 50	13	55.2	0.42–1.85	1.11±0.12	21	57.3	0.34–3.15	1.33±0.09
Totals	61	40.2	0.29–2.09	0.95±0.05	60	46.6	0.20–3.15	1.14±0.06

*Mean ± standard error of mean.

*It has been shown previously[4] that specimens collected at 9 A.M., 11 A.M., 1 P.M., and 3 P.M., are essentially the same, the only variation occurring in the 1 P.M., specimen, which was slightly higher than the others.

Since it has been shown that the degree of erosion increases with age, the mean for the erosion cases was corrected for age by analysis of covariance.[39] The difference in the means of erosion and normal cases was then found to be not statistically significant.

A wide range in values for citric acid in saliva is evident in the data appearing in Table II as well as in Figs. 6 and 7. Based on comparatively small groups, the data are inconclusive as regards the relation between citric acid and erosion. However, some indication of a positive trend in the data is apparent by the magnitude of the correlation coefficient, as shown in Fig. 7.

A scatter diagram and line of regression relating age to salivary citrate is shown in Fig. 6.

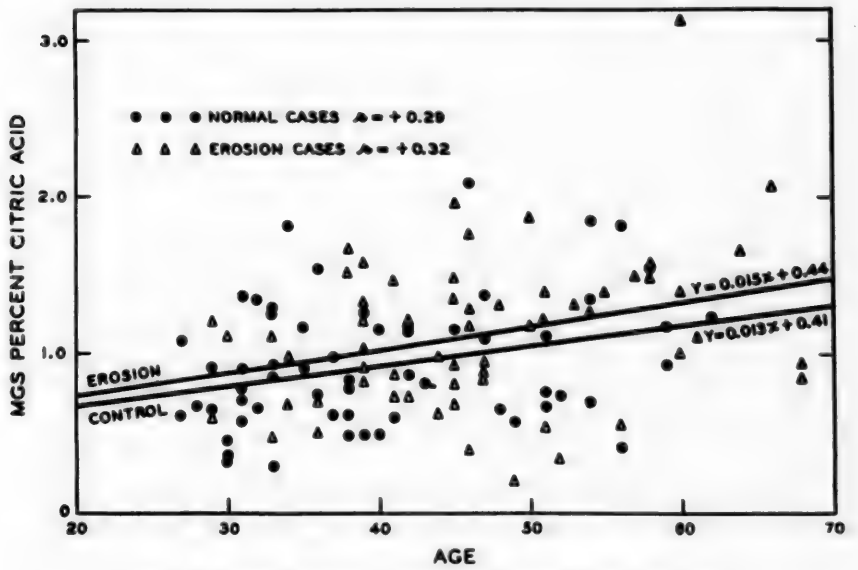

Fig. 6.—Relation between citric acid content of saliva and age.

The coefficient ($r = +0.29$) for normal individuals, indicates a result happening by chance less than 5 times in 100 trials. The coefficient $r = +0.32$ for erosion cases could have occurred by chance less than twice in 100 trials. A prediction that there is an increased content of citric acid in saliva of older age groups is warranted by this comparison. (In one erosion case a value of 3.15 mg. per cent was obtained. This analysis was repeated five months later and essentially the same result was obtained.)

The correlation coefficient ($r = +0.33$) associated with the comparison of citric acid and erosion index, as shown in Fig. 7, has a probability of occurrence by chance of less than once in 100 trials.

Inspection of the results as presented in Figs. 6 and 7, therefore, brought out significant correlations between citric acid content and two independent

variables, that is, age and erosion index. Under this circumstance, a treatment of the data by means of multiple correlation was desirable and this is shown in Figs. 8 and 9.

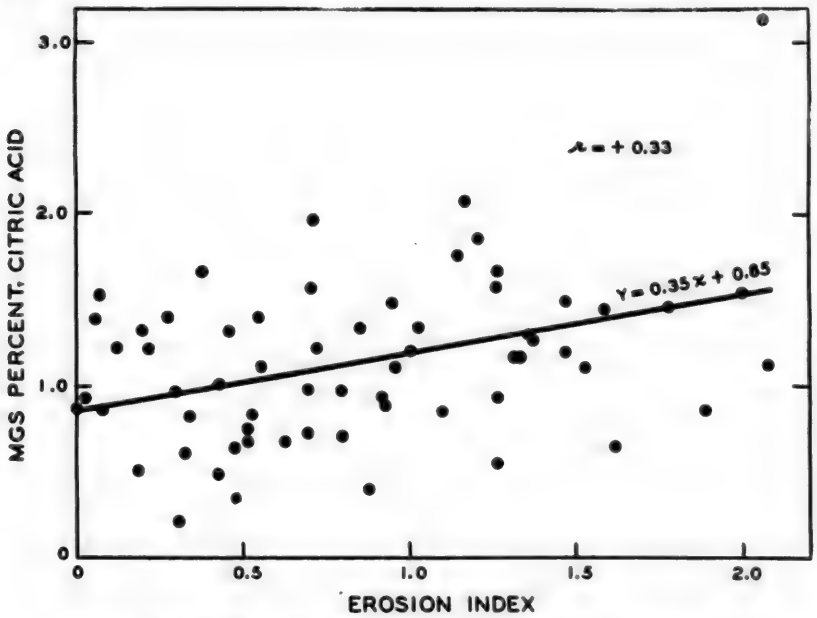

Fig. 7.—Relation between citric acid content of saliva and erosion index.

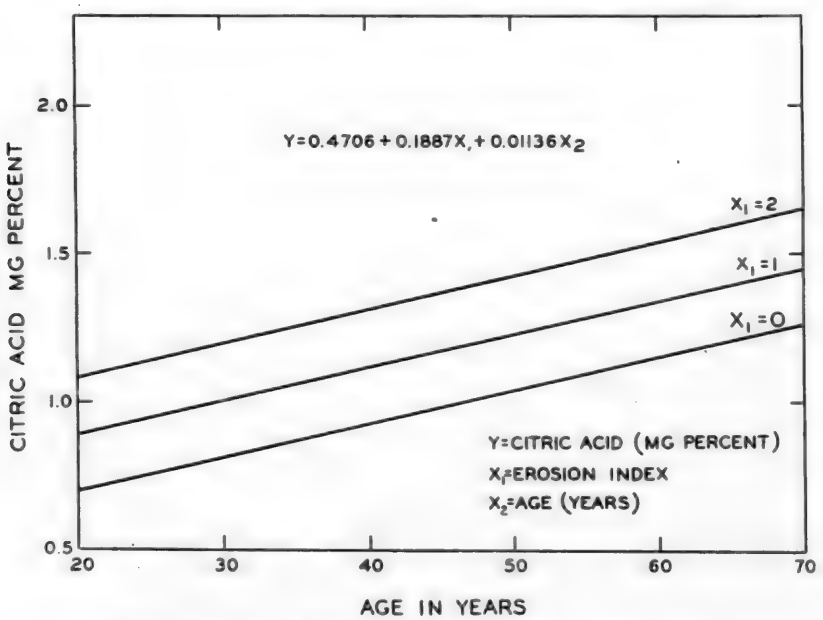

Fig. 8.—Relation of citric acid to age at constant erosion index.

These regression coefficients were calculated by use of the following simultaneous equations:

$$An + B\Sigma X_1 + C\Sigma X_2 = \Sigma Y$$
$$A\Sigma X_1 + B\Sigma X_1^2 + C\Sigma X_1X_2 = \Sigma X_1Y$$
$$A\Sigma X_2 + B\Sigma X_1X_2 + C\Sigma X_2^2 = \Sigma X_2Y$$

where A, B, and C are the regression coefficients, X_1 = erosion index, X_2 = age in years, Y = citric acid content in mg. per cent and n = 121. These regression coefficients were tested for significance according to Fisher,[39] and both were found to be statistically significant.*

The correlation between the erosion index and the citrate content of saliva at any given age is expressed in Fig. 9.

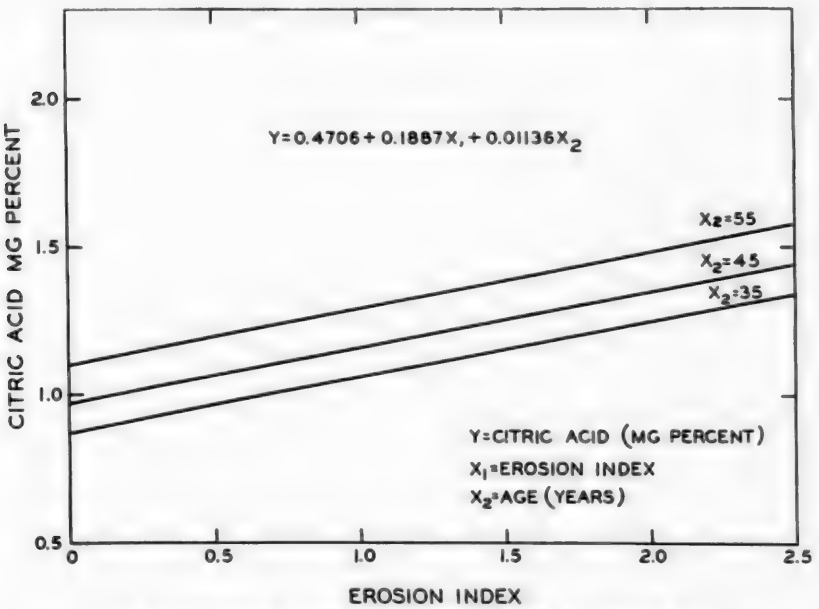

Fig. 9.—Relation of citric acid to erosion index at constant age.

This correlation, which is more useful than the correlation shown in Fig. 7, since the latter does not take into account the effect of age, suggests a relation between the severity of erosion and the citrate content of saliva. However, it has been shown that the mean salivary citrate content of the normal individuals does not differ significantly from that of the erosion cases when corrected for age. It appears at first glance that these two findings are not in accord. It should be remembered, however, that the mean citrate content for the erosion cases is somewhat higher than that for the normal cases. Moreover, it is to be noted that individual values comprising the mean for the erosion cases show a positive trend (Fig. 7). Hence, even when corrected for age, a correlation may exist between the severity of erosion and the salivary citrate content (Fig. 9).

*We are indebted to Mr. Jerome Cornfield, Statistician, Division of Public Health Methods, Office of The Surgeon General, USPHS, for statistical treatment of the data.

DISCUSSION

The prevalence of erosion among our subjects would appear to be unexpectedly high, particularly by comparison with the recent study by Shulman and Robinson[9] who found gross evidence of erosion in only 2 per cent of a group of 1,345 male college freshmen. In all probability, these were individuals under the age of 30 years. Kitchin[40] reported the following per cent prevalence of abrasion due to toothbrush and dentrifice: age 20 to 29, 42 per cent; age 30 to 39, 45 per cent; age 40 to 49, 76 per cent, and age 50 to 59, 70 per cent. It has been postulated by Souder and Schoonover[41] that slightly abrasive dentrifices may damage the enamel by virtue of the solvent action of a chemical ingredient, such as sodium metaphosphate, present in some dentrifices.

It was our experience, and this has been the experience of other investigators,[42] that it is extremely difficult to make a clear-cut differentiation between loss of tooth substance due to mechanical factors and that due to purely chemical agents. For purposes of this study, as noted above, erosion was diagnosed as a loss of tooth substance on the gingival third of the tooth on the labial or buccal surfaces, regardless of cause. Shulman and Robinson,[9] however, state in their report that "Erosion was differentiated from dentrifice abrasion by its position, and from dental caries by the quality of its base."

The action of citrate on rats' oral tooth surfaces, noted in a previous study,[12] resembles the gross appearance of human erosion and previously suggested "that saliva (or localized fluid exudates bathing tooth surfaces) may contain citrate or other anions which effect calcium solubility and advance the destruction of tooth surfaces orally."[12] The possibility arises that specimens of stimulated saliva represent a dilution of the localized diffuse oral secretions to such an extent that salivary citrate analyses fail to reveal the full magnitude of differences in erosion and nonerosion individuals. The analytical differences as we have reported, therefore, may possibly possess even more significance than is apparent in the statistical analyses of the data.

The hypothesis that human dental erosion may be a localized nonacid decalcification due to a calcium solubilizing anion such as citrate, while not clearly demonstrated in the results of this clinical study perhaps, should not be discarded as a good working hypothesis for future study. On the basis of the present data, it can be said that the severity of human erosion is associated with the salivary citrate content. In view of the limited number of cases it is felt that more work should be done to corroborate these findings.

Our results vary somewhat from a recent report by Shulman and Robinson,[12] although it must be noted that the age groups differed in these two studies, as well as the technic employed in preserving and analyzing the saliva specimens. In this connection it may be noted again that the preservation of saliva prior to analysis for citric acid is an extremely important factor.

SUMMARY

1. Application of the citric acid method proposed by Pucher, Sherman, and Vickery and modified by Perlman, Lardy, and Johnson, for the analysis of

saliva, was critically studied and a satisfactory procedure adopted. The citric acid in saliva is quite unstable, but by addition of H_2SO_4 the saliva is stabilized and recovery of added citric acid is satisfactory. Amounts of citric acid varying from 0.010 mg. to 0.450 mg. in 10.0 ml. specimens of saliva may be determined.

2. Although the number of individuals involved is small, the general pattern of erosion was observed to be as follows: (a) the upper teeth show a greater degree and a greater prevalence of erosion than the lower teeth, (b) the teeth of the right and left quadrants of both jaws have a similar degree of erosion, and (c) the severity of erosion increases with age.

3. The citric acid content of stimulated saliva varies from 0.20 mg. per cent to about 2.00 mg. per cent in individuals with and without dental erosion. There is a slight increase with age in the citric acid content of stimulated salivas.

4. On the basis of the data thus far accumulated, there appears to be a positive statistical correlation between the severity of human erosion and the salivary citrate content.

We gratefully acknowledge the cooperation of the following in making erosion cases available to us for examination and for collection of saliva: (1) Bruce D. Forsyth, Senior Dental Surgeon, USPHS, USPHS Dispensary, Washington, D. C.; now Assistant Surgeon General (Dental), USPHS. (2) A. E. Nannestad, Dental Director, USPHS, Marine Hospital, Baltimore, Md.; now at the U. S. Marine Hospital, Philadelphia, Pa. (3) R. S. Lloyd, Senior Dental Surgeon, USPHS, Marine Hospital, Baltimore, Md. (4) E. G. Pollard, Commander, USN, Navy Medical Center, Bethesda, Md.

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INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Proceedings of Twenty-Seventh General Meeting

Drake Hotel, Chicago, Ill.

June 24, 25, and 26, 1949*

Compiled by Hamilton B. G. Robinson, Editor, *College of Dentistry, Ohio State University, Columbus, Ohio*

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I. Inaugural Address: Dental Education and the International Association for Dental Research

J. R. Blayney, Chicago, Ill.

It appears to have become the custom for the incoming President to discuss some topic of general interest. One year ago the Secretary called to my attention the fact that I would be expected to present such a discussion at this time. I was told it was customary to speak regarding one's own investigations or some topic of historical interest. In reviewing the titles of the addresses given by my predecessors I have found that they extend from a critical analysis of research or problems of dental educational to that interesting and entertaining presentation of "Operations Crossroads." Being neither an atomic scientist nor dean of a progressive dental school I cannot hope to present for your consideration the devastating effects of modern warfare or a model curriculum for undergraduate dental education.

Before entering upon my discussion I should like to gratefully acknowledge and to express my appreciation for the high honor you have given me. I accept this office with a realization of the many responsibilities involved because I firmly believe that men of your character and interest will wholeheartedly assist in advancing the activities and success of the association. Without this cooperation my efforts will have little value.

While reflecting upon the various topics which would be appropriate to discuss at this time it occurred to me that it should be of interest to pause for a few moments to consider what progress our association has made and what has been its influence on dental education. I will speak briefly on the topic: "Dental Education and the International Association for Dental Research."

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*The meetings of the International Association for Dental Research preceded the meetings of the American Association of Dental Schools on June 27, 28 and 29.

The Editor prepared these Proceedings with the close cooperation of the General Secretary, Dr. Edward H. Hatton.

We have celebrated the one hundredth anniversary of the founding of the first dental school. Much has been accomplished during the first century of dental education. A great deal more remains to be achieved. From a humble beginning, but under the guidance of sincere and far-seeing leaders, dentistry has advanced from the level of a craft to that of a full rank in the healing arts professions.

Five very important milestones stand out when we review the history of dental education. They are:

1. The organization of the *Journal of Dental Research* in 1919.
2. The organization of the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH in 1920.
3. The study of dental education by the Dental Educational Council with the report prepared by Dr. Gies entitled "Dental Education in the United States and Canada in 1921 to 1926."
4. The report of the Curriculum Survey Committee of the American Association of Dental Schools in 1935.
5. The formation of the Council on Dental Education by the American Dental Association in 1937 and its study of the dental schools from 1942 to 1945.

Of course other achievements of merit can be mentioned but from our present vantage point those cited stand out in bold relief. For our purpose today I believe we can consider item one, the organization of the *Journal of Dental Research,* and item two, the organization of our ASSOCIATION as being one and the same as they are so closely knit and related.

The INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH was founded in 1920 with twenty-five charter members in New York City, fourteen in Boston, and one in Chicago. By the end of 1920 there was a nucleus of three sections with fifty-three members since by that time the Chicago group numbered fourteen. The objectives as announced were to advance research in all branches of dental science, to encourage and facilitate cooperative efforts and achievements among investigators so as to promote dentistry for a more perfect service to humanity. From 1920 through 1927 three more sections were established, namely, Toronto in 1921 with a membership of fourteen, Ann Arbor in 1923 with five, and San Francisco in 1924 with seven members. Also seven memberships were issued to those living in areas where a local group was not active. During those eight years, twelve memberships were lost either through death or resignation. As of January 1, 1928, the ASSOCIATION had a total membership of seventy-four. During this year (1928) seventy-three memberships were issued with sections appearing in several centers. Without taking time to trace the growth year by year the ASSOCIATION attained a membership of five hundred and nineteen in twenty-six North American sections and with ninety-one members in seven sections in foreign lands at the close of 1948. While we may point with pride at this excellent record of growth in physical stature there are other criteria which are much more indicative.

The report published in 1926 by Dr. Gies in the *Bulletin on Dental Education in the United States and Canada* reveals that research in dental educational institutions was at a very low ebb. Of the forty-six institutions studied by the committee we find that only nine schools indicated that they had what was called a research program. The budgets varied from a low of $50.00 to a high of $15,073.00 with a median of $1,773.00 for the academic year 1920-1921. During the year 1923-1924 fourteen schools had research programs with budgets running from $71.00 to $34,821.00 with a median of $2,000.00. It is difficult to draw a

definite comparison with the findings of the Council on Dental Education's study of 1942-1943, inasmuch as the item on research in the latest study did not request such budgetary figures, or a statement of the programs in progress. However, all of the thirty-eight schools studied by the Council indicated that they did have a research program. Probably the least of these would fully equal the median program of 1920-1924. It is of course well recognized that many of our institutions had research programs which the Council considered as very effective from the standpoint of physical facilities, allocation of funds, and the results obtained.

It is entirely fitting that we ask ourselves the questions: What is the place of a research program in a dental school? Does the responsibility of a dental school go beyond that of training a student to practice his profession? It is well accepted that the objective of higher education is not limited to the dissemination of factual material. The objective includes inquiry into the vast and unexplored areas of knowledge. Teaching and research in our professional schools go hand in hand. President Stoddard has said, "The aim of higher education is not to turn out 'educated' men and women emerging bright and shiny like so many insects from a four-year cocoon. The aim is rather to give students a living fund of knowledge from which they may generate ideas. The aim is to encourage straight thinking."

Research is an orderly planned and executed endeavor to explore the unknown and to extend the breadth of knowledge. Without such a program stagnation exists and teaching becomes stifled. Education at the university level is more than the dissemination of knowledge. The search for new truths is just as important. Without the stimulating influence of research professional education becomes hollow and soon adopts empirical methods. Before 1920 research in dentistry was chiefly directed towards the development of new appliances or new technical procedures primarily directed toward the repair of dental defects. A major portion of this was under the direction of commercial concerns.

Probably because of this technical approach rather than the study of biological principles underlying the etiology of oral disease, dental education at this period stressed reparative methods in lieu of prevention. Departments within an institution which foster an active investigative program exhibit the most enthusiastic and effective teaching. This type of teaching will stimulate the students of the higher level to develop the spirit of enquiry and thereby do much to encourage them to seek an understanding of the scientific principles which will unlock the door to prevention as well as a more rational method of therapy. A dental curriculum which does not include an active investigative program fails to provide the proper stimulus to the faculty for more effective teaching and to the student body for the development of enquiry regarding the conditions with which they will be confronted.

In the President's address of 1928, Dr. Miner pointed out that three groups were primarily concerned in the problem of dental research, namely the investigators, the administrators, and the donors. It has been the general belief that research on dental or oral problems was lagging because of the almost total absence of financial support. Little attention was given to the thought that dental education had failed to impress either the administrators of the university graduate and research programs or the general public that there was a crying need for investigation concerning oral disease. Also it was considered that dental education itself was not striving to create a spirit of enquiry among the more promising students. With this apparent lack of interest on the part of dental education it is not surprising that funds were not forthcoming to promote basic scientific investigation regarding oral disease.

With the establishment of the INTERNATIONAL ASSOCIATION OF DENTAL RESEARCH the first step was taken on the part of dentistry to put its own house in order. When the stronger schools within universities began to demonstrate the ability of some of their faculty members to conduct research, university funds for the promotion of research gradually became available. As those studies contributed to a better understanding of the problems of oral disease more and more funds were forthcoming both through university budgets and from private sources. It is true that dental disease is not dramatic when compared with cancer or poliomyelitis; however, private funds can be obtained when the public understands the need. Mr. Walter G. Zoller was one of dentistry's greatest benefactors. It was a practicing physician who was intensely interested in the role which oral conditions play in systemic disease who first interested Mr. Zoller in dentistry. During a recent visit to the Zoller Clinic, Mr. Goldblatt, who has given so much money and now is devoting all of his time to the raising of funds for cancer research asked me, "Are you telling the public what you are doing? If you will tell them the need as you have told me I can get you all the money you will use. It is easy to get money but you must first tell the public."

I am convinced that Mr. Goldblatt is right. When we have developed the personnel capable of conducting scientific investigations yielding results which are comparable and as significant as research in other areas of science, we will enjoy an equal distribution of financial aid.

In the report of the President's Scientific Research Board on "The Nation's Medical Research" mention was made of the acute shortage of adequately trained personnel for dental research. Dr. H. Trendley Dean has indicated that only one-half of the existing dental schools have made application for financial assistance for investigation from the National Institute of Health Grant-in-Aids. The principal reason for this is the shortage of personnel.

In order that our more recent members may better understand the role that the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH has played in the development of research in the dental schools, I should like to refer briefly to the early days of the ASSOCIATION. The first annual meeting was held in New York City on December 21, 1922. Two papers were presented at that time; one by C. F. Bodecker on "The Dento-Enamel Circulation" and one by Dr. J. Leon Williams on "Permeability of Enamel." The second meeting was held in Chicago on March 7, 1924. Four presentations are recorded for that session. One consisted of a presentation of old and rare prints and etchings depicting the history of dental practice, two were concerned with histology and pathology of oral structures, and one discussed the presence of fusiform bacilli and spirochetes in alveolar abscesses. The third annual meeting occurred on March 20, 1925, in Chicago at which five reports were made. The fifth meeting was held in New York City on April 19, 1927, with four presentations. No mention was made of the number in attendance at these meetings. The sixth annual meeting was held in Washington, D. C. on March 25, 26, 27, and 28, 1928. The total enrollment as of March 15, 1928, was eighty-one members. While this meeting was distributed over four days, in reality it consisted of five sessions. These sessions were so arranged so as not to interfere with the fifth annual meeting of the American Association of Dental Schools. During these five sessions forty-seven reports were presented. It was during this annual meeting that the seventy-three new names were added to the official roster. The total registration of the meeting was forty-seven. In reviewing the reports which were presented for discussion we find only a limited number which today could be considered as research reports. Many would have to be classed as case reports or opinions of the authors which were not substantiated by convincing data.

This is in striking contrast with the twenty-sixth annual meeting held in Rochester, New York, on June 18, 19, and 20, 1948. At this meeting there were five sessions and on two occasions the session was divided into two sections. The attendance of members was one hundred thirty-eight with an addition of one hundred thirty-two guests. One hundred and four reports were presented and discussed while an additional fifty-nine reports were read by title. A large percentage of these one hundred sixty-three research reports were based upon well-organized and carefully conducted experimental study.

Dr. Hatton has made a comparison of the areas covered by the research reports presented in the 1928 and 1929 meetings with those presented at the 1948 meeting. (Table I.) These show a very interesting and, I believe, significant trend.

TABLE I

SUBJECT AREA	1928-1929 (%)	1948 (%)
Anatomical	17.5	2.5
Bacteriology and parasitology	15.0	5.0
Chemistry and pharmacology	5.7	13.5
Clinical research	13.5	6.8
Clinical research in caries prevention	0.0	5.0
Growth and development	0.0	9.5
Histology and embryology	14.5	4.3
Metallurgy, etc., general program	1.0	1.3
Metallurgy, etc., dental materials group	0.0	14.5
Non-research	11.7	5.0
Nutrition and diet	9.0	8.5
Pathology, tumors, pathology of dental caries	4.7	17.5
Physiology	4.7	4.3
Physics and physical chemistry	0.0	3.0

The motivation for the spirit of enquiry in our dental educational institutions so admirably initiated by the founders of the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH and advanced by the enthusiastic support of the entire membership has done much to elevate the standing of dental education to the level of a university discipline. That much remains to be accomplished regarding the development of research programs in our dental schools is borne out by the fact that only eighteen of the dental schools in the United States supplied items to the 1948 program. Only in very recent years have our educational institutions begun to consider the preventive phase of dental practice. It is evident that dental education must place major emphasis upon the study of the child. These achievements will be accomplished through intensive and carefully planned investigation.

In the past twenty-nine years our ASSOCIATION has contributed magnificiently to the general welfare of the public. We have assisted materially in the development of dental education to the position which it now enjoys. The stimulation of the spirit of enquiry has always been encouraged and will be continued at an ever increasing tempo. This has been a very important factor in the organization and development of what is truly a graduate study. The INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH has made significant contribution to the advancement of dentistry. For this we may be justly proud.

The record reveals that the ASSOCIATION has lived up to the announced objective to advance research in all branches of dental science, to encourage and facilitate cooperative efforts and achievements among investigators so as to promote dentistry for a more perfect service to humanity.

II. Index of Participants, and Sequence Numerals for Corresponding Abstracts

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III. First Session: Friday Afternoon, June 24; Abstracts 1-13

1. The Effect of Water Soluble Chlorophyll "A" on Lactobacillus Acidophilus Counts of Saliva. *G. W. Rapp, Ph.D., Loyola University School of Dentistry, Chicago, Ill.*

2. The Effect of Water Soluble Chlorophyll "A" on Certain Enzymic Activities of Human Saliva. *B. F. Gurney, M.S., and G. W. Rapp, Ph.D., Loyola University School of Dentistry, Chicago, Ill.*

3. The Effect of Water Soluble Chlorophyll "A" on Mouth Odors. *G. W. Rapp, Ph.D., and Albert A. Olen, B.S., Loyola University School of Dentistry, Chicago, Ill.*

4. The Effect of Sodium Copper Chlorophyllin on Experimental Caries and on In Vitro Acid Production in Incubated Saliva-Carbohydrate Mixtures. *William G. Shafer and John W. Hein, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.*

5. The Effect of a Single Exposure to X-Ray Irradiation on the Growth Rate of Dentin of the Rat Incisor. *M. Weinreb, I. Schour, H. Medak, and W. A. Klauber, Jr., Schools of Dentistry and Medicine, University of Illinois, Chicago, Ill.* This study is based on the investigation of 22 albino rats, ranging in weight at the beginning of the experiment from 150 to 200 grams. Fourteen animals were exposed to a single irradiation of 4000 r upon the area of the upper rat incisor, the remainder of the body being protected. Eight animals served as control. The x-ray machine was standardized at 100 kv., 5 Ma., t.s.d. (target-skin distance) of 12½ cm., no filter, and H.V.L. of 1 mm. Al (aluminum). The size of the field in all doses was a circle with diameter of 2.5 cm. The output was 320 r per minute. The experimental animals were given intraperitoneal injections of Alizarin red sodium salt one week prior to the date of irradiation, immediately following exposure, and at weekly intervals thereafter. They were sacrificed at various intervals after irradiation, ranging from 1 to 21 days. The upper incisors were embedded in plastic medium and ground sections were prepared. The weekly growth of dentin in width was measured between successive alizarin lines. The weekly growth of dentin in length was measured between the points of emergence of the alizarin lines at the dentino-enamel junction. The growth of dentin in length was stopped abruptly following irradiation. The growth of dentin in width continued undisturbed. Eruption slowed down and eventually stopped, but the decline was much more gradual. The length of the clinical crown decreased continuously by attrition until the crown was almost worn to the gingival margin.

6. The Effect of X-Ray Irradiation on the Eruption of the Upper Incisor of the Albino Rat. *H. Medak, I. Schour, and W. A. Klauber, Jr., Schools of Dentistry and Medicine, University of Illinois, Chicago, Ill.* The effects of a single exposure to x-ray on the eruption of the upper incisor was studied in 80 white rats weighing 180 to 200 grams. Eight were irradiated with 500 r; 8 with 1000 r; 8 with 2000 r; 8 with 2500 r; 12 with 3000 r; and 28 with 4000 r units. Eight animals were used as control. The animals were observed up to 20 weeks. The animals were anesthetized with Nembutal. Only the area of the upper incisors was exposed to irradiation. The x-ray machine was standardized at 100 kv., 5 Ma., t.s.d. of 12½ cm., no filter, and H.V.L. of 1 mm. Al. The size of the field in all doses was a circle with diameter of 2.5 cm. The output was 320 r per minute. The rate of eruption was determined by notching the upper incisor at the gingival margin. The eruption rate of the upper incisor of

the normal controls was 2.0 to 2.6 mm. per week. The animals receiving 500 and 1000 r showed a similar curve. The animals receiving 2000 r showed a dip in the eruption rate for the first 6 to 7 weeks and from then on a normal curve. Groups receiving 3000 r and 4000 r showed within three weeks a drop in the rate of eruption to between 0.2 to 0.4 mm. per week. The rate remained between 0.0 and 0.4 mm. until the end of the period of observation. Histologic studies will be made to trace the mechanism of the striking retardation and cessation of eruption.

7. The P-M-A Index of Gingivitis. *Maury Massler and Isaac Schour, School of Dentistry, University of Illinois, Chicago, Ill.* The P-M-A method of quantitatively assessing gingival disease is based upon the examination of each individual gingival unit in the mouth (the papillary [P], marginal [M], and attached [A] gingiva mesial to any given tooth). This method offers many advantages over previous subjective clinical methods and has been applied during the past two years to a total of approximately 10,000 persons ranging in age from 2 to 60 years. Gingivitis and recession were assessed separately. The gingivitis attack rate was characterized by two phases. The first consisted of a very rapid rise from a null point (0.0 per cent affected) at 5 years to a peak at 11 to 13 years (80 per cent affected). A sharp decrease to a young adult level occurred at about 16 years of age (60 per cent affected). There was no apparent increase in gingivitis between 16 and 25 years. The second phase began at approximately 25 years of age. It increased very slowly in contrast to the first phase and followed a simple cumulative pattern. During the second phase, two distinct rates of attacks could be distinguished: A higher incidence of gingivitis in a group of factory workers (80 per cent affected), and a lower one in a group of office workers (65 per cent affected). The ten anterior gingival units (upper and lower anterior segments) bear the brunt of the majority of attacks of gingivitis. This fact suggested that the P-M-A examination might be restricted to the anterior segments, just as D.M.F. surveys are often restricted to the examination of the first permanent molars alone.

8. Bacterial Antagonisms and Dental Caries. *Charles A. Scrivener, D.D.S., Hugh I. Myers, Ph.D., Norman A. Moore, D.D.S., Ben W. Warner, D.D.S., M.S.D., The University of Kansas City, School of Dentistry, Kansas City, Mo.* In an earlier publication Schantz and Scrivener reported that the rate of dissolution of tooth fragments in saliva from caries-resistant persons was greater than that from active caries saliva. This result was contrary to what was expected and preliminary evidence indicated antibiosis could be the explanation. The present work was initiated to confirm the previous data and gather additional proof, if possible for the antibiosis hypothesis. Ten dentin fragments were weighed and with the proper media and bacteria placed in a sample bottle and incubated. Suitable additional weighings were recorded. To establish the curve of weight loss for tooth fragments, a series of experiments were performed using tooth fragments and distilled water. It was found that, although variable, the weight loss was greatest initially and then became more gradual. Variation between bottles containing tooth fragments from the same tooth was less than that for bottles with fragments of different teeth. Five control experiments showed a weight loss. In the attempt to reisolate from saliva an unnamed streptobacillus reported in the earlier publication, an organism with some of the same characteristics was found. This organism did not prevent tooth fragment weight loss in nutrient broth, but in nutrient broth, tooth fragments, lactobacillus, and yeast, it caused

a weight gain. In similar experiments with *Streptomyces griseus* and yeast, and *Streptomyces lavendulae* a similar weight gain was recorded. Thus some additional evidence was obtained concerning the antibiosis involved in the caries process.

9. Bacillus Brevis as a Dental Caries Bacterial Antagonist. *Charles A. Scrivener, D.D.S., Hugh I. Myers, Ph.D., Norman A. Moore, D.D.S., and Ben W. Warner, D.D.S., M.S.D., The University of Kansas City, School of Dentistry, Kansas City, Mo.* Evidence from Schantz and Scrivener and preliminary experiments on three other bacteria gave some evidence of antibiosis as a possible factor in dental caries. This report concerns experiments on *Bacillus brevis.* A mixture of *B. brevis, Actinomyces, lavendulae, Actinomyces griseus,* and streptobacillus added to a culture of nutrient broth, tooth fragments, and lactobacillus caused a four-hour gain in weight. In comparison, several control experiments showed a loss in weight. Four additional series of experiments with broth, tooth fragments, sucrose, lactobacillus, and *B. brevis* confirmed the gain in weight. Bacteriologically *B. brevis* prevented bromcresol green agar inoculated with saliva from giving the acid reaction. *B. brevis* also prevented lactobacillus growth on tomato juice agar plates. The dialysate from the dialysis of brevis cultures inhibited the growth of lactobacillus on smear tomato agar plates. In the proper concentrations, sodium fluoride was found to stimulate *B. brevis* growth. Hence, *B. brevis* and some other organisms have been found to possess properties that are inhibitory to the processes of dental caries from the standpoint of physical, chemical, and bacterial activity in vitro.

10. The Evanston Dental Caries Study. V. The Fluorine Content of Saliva and Its Relationship to (A) Oral Lactobacillus Counts and (B) the Prevalence of Dental Caries. *D. J. Martin and I. N. Hill, Evanston Health Department and Walter G. Zoller Memorial Dental Clinic, Evanston and Chicago, Ill.* As a part of the Evanston Dental Caries Study the following investigations have been made: (1) The analysis of saliva to determine its fluorine content; (2) the relationship between the fluorine content of saliva and (A) the oral lactobacillus count, and (B) the prevalence of dental caries. The stimulated salivary specimens were collected on the Monday of each school week from 10 per cent of the children in the Dental Caries Study. The specimens were collected for a period of one school year before either the clinical or roentgenological examinations had been made. The time interval of exposure to sodium fluoride, which has been added to the Evanston water supply since February, 1947, varied in the two groups studied. The 6-7-8-year age group was exposed to the fluorided water from 20 to 25 months and the 12-13-14-year age group from 25 to 29 months. In the 6-7-8-year age group the fluorine content of the saliva varied from 0.14 p.p.m. to 0.35 p.p.m. with an average of 0.25 p.p.m. and a mean of 0.27 p.p.m. In the 12-13-14-year age group the fluorine content of the saliva varied from 0.11 p.p.m. to 0.29 p.p.m. with an average of 0.20 p.p.m. and a mean of 0.20 p.p.m. The results of this investigation show no apparent relationship of the fluorine content of saliva to the oral lactobacillus counts or to the prevalence of dental caries.

11. Evaluation of Disinfectants: A Report of Infection-Prevention Tests of Some Common Disinfectants. *Marjorie P. Prombo and Evelyn B. Tilden, Department of Bacteriology, Northwestern University Dental School, Chicago, Ill.* There were striking differences in the killing power of various disinfectants according to what test organism was used. Iodine and saponated cresol were almost completely effective against all three of the bacteria used in

the tests. The 3 per cent formaldehyde solution of Tainter et al., and 1 per cent tincture of the quaternary ammonium compound, DC 12, killed both pneumococcus and tubercle bacillus and were reasonably active against hemolytic streptococcus, but DC 12 in aqueous solution was useless against tubercle bacillus. Zephiran chloride behaved much like DC 12 against pyogenic cocci. It was necessary to use these two quaternary compounds in relatively high concentration (1 per cent). Compounds of this type are not necessarily bactericidal. For example, cetylmethylethylamminoum bromide (Radiol) appeared to be inactive when used in the concentration recommended by the makers. Phenol and the phenol derivative, orthophenyl phenol, failed against streptococcus, though active against both tubercle bacillus and pneumococcus. Although tubercle bacillus is usually thought to be harder to kill than streptococci or pneumococci, because of its resistance to acids, alkalies, chlorine, and mercurials, it is noteworthy that in the case of saponated cresol a solution of 2 parts in 100 was sufficient to kill tubercle bacillus, while 5 parts in 100 was necessary for two pyogenic cocci. On the basis of these tests, it would seem that a safe choice for disinfection of dental instruments would be 5 per cent saponated cresol or the 3 per cent formaldehyde solution of Tainter and his associates. A 1 per cent tincture of DC 12 would be nearly as good, though not quite so effective against hemolytic streptococcus, while Metaphen disinfecting solution, which contains benzyl alcohol, would be highly satisfactory for hemolytic streptococcus. Aqueous solutions of Metaphen and Merthiolate should certainly not be relied upon, and orthophenyl phenol would seem to be of doubtful value, because it lacks ability to kill hemolytic streptococcus, even though it is very effective against the other two bacteria, especially tubercle bacillus.

12. The Effect of Fluoride on the Solubility of Human Enamel. *Frank J. McClure and Robert C. Likins, National Institute of Dental Research, National Institutes of Health, Bethesda, Md.* Determinations have been made of the solubilities of nonfluorosed, and NaF treated human enamel in eight acid solutions. All samples were obtained from pooled aggregates of six or more teeth, powdered to 60 mesh, and separated after the Manly-Hodge technic. Fluoride analyses, expressed as per cent of enamel ash, were: nonfluorosed—0.0111; fluorosed—mild—0.0618, moderate—0.0909, severe—0.0758; NaF treated—0.0460. Next, 0.01N solutions of formic lactic, acetic, tartaric, citric, hydrochloric, nitric, and phosphoric acids were prepared. Fifty milligram samples of enamel were stirred continuously in 25.0 c.c. of acid at 37.0° C. for 17 minutes. The suspension was centrifuged and an aliquot of the supernatant removed at T_0 + 20 minutes. The supernatant liquids were analyzed for Ca and P according to standard procedures. Comparative solubilities were expressed in terms of per cent Ca and P dissolved. The data indicate that fluorosed and NaF treated enamel were less soluble in formic, lactic, acetic, tartaric, and citric acids than nonfluorosed enamel. Differences in solubility in hydrochloric, nitric, and phosphoric acids were not observed.

13. The In Vitro Adsorption of Fluoride by Human Enamel. *R. C. Likins and F. J. McClure, National Institute of Dental Research, National Institutes of Health, Bethesda, Md.* The adsorption of fluoride by powdered enamel from solutions of ten different fluoride salts has been determined. Each solution contained 8 p.p.m. fluorine. The enamel samples used were 160 mg. aliquots of a pooled aggregate of approximately 300 sound teeth, powdered to 60 mesh, and separated by the Manly-Hodge technic. Samples were stirred for 5 minutes in 40 ml. of fluoride solution at 37° C., centrifuged, and the solution decanted. Washing of the enamel sample was accomplished by resus-

pending in 20 ml. of distilled H_2O, centrifuging, and decanting. Samples were studied in quadruplicate; two samples were washed 5 times and two, 10 times. The enamel samples were then ashed and analyzed for fluorine. Significant variations in the fluoride adsorbed by the enamel from the different solutions were observed. The data also suggest that adsorbed fluoride is removed by the washing process.

IV. Second Session: Friday Evening, June 24; Abstracts 14-22

14. A Comparative Study of the Efficiency of 2 Per Cent Procaine With and Without Epinephrine 1:50,000. *Emmett R. Costich, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* A study was undertaken with 50 medical students ranging in age from 22 to 32 years as experimental subjects. Each student received five right mandibular block injections at weekly intervals. The initial injections served only as training and were disregarded in tabulating data. The remaining four consisted of two injections of 2 c.c. of each type solution. It was found that plain procaine gave a slightly faster onset of anesthesia but had a mean duration of only 36 minutes as compared to 90 minutes for procaine-epinephrine. The only significant difference noted in pre- and postinjection recording of pulse, blood pressure, and respiration was a drop in the diastolic blood pressure following procaine-epinephrine. There was a significantly greater incidence of systemic reactions of tremor, sweating, and faintness and postanesthetic stiffness following the procaine-epinephrine. The quality of anesthesia with procaine alone was much less profound as compared to the procaine-epinephrine solution.

15. Comparative Studies of Streptomycin and Penicillin in the Treatment of Infections of the Oral Mucosa. *S. Leonard Rosenthal, D.D.S., School of Dentistry, Temple University and Gordon R. Winter, D.D.S., School of Dentistry, University of Pennsylvania, Philadelphia, Pa.* The concentrations employed were: penicillin G 10000 Oxford units per c.c.; streptomycin 100 mg. per c.c. of water. Studies in vitro demonstrated the effectiveness of both antibiotics. Equal parts of fresh saliva and either antibiotic in contact for 15 minutes sufficed to prevent all growth in aerobic and anaerobic cultures. When loss of motility was the criterion, streptomycin penetrated the organic and inorganic contents of saliva more readily than penicillin. Clinically, both gave satisfactory results; but streptomycin acted more rapidly in relieving symptoms, reducing bacterial numbers, and stopping motility of the motile flora. Streptomycin seems superior for topical application, penicillin for parenteral use.

16. Factors Influencing the Incidence of Bacteremias Following Surgical Procedures in the Oral Cavity. *Joseph P. Lazansky, Leonard Robinson, and Lillian Rodofsky, Tufts College Dental School, Boston, Mass.* The incidence of transient bacteremias following tooth extraction and periodontal scalings was studied in 221 cases. In the initial 161 cases, 10 c.c. samples of blood were taken prior to operation, and at one and 10 minutes following the operation. In the remainder of cases, the preoperative sample was routinely omitted, the first postoperative sample increased in volume to 20 c.c. and second postoperative sample taken at 5 minutes. Transient bacteremias were detected 22 times in 221 operations, an over-all incidence of 10 per cent. The organisms were alpha hemolytic streptococci, except in one instance in which

the organism was a nonhemolytic (gamma) streptococcus. In 56 single extractions, one positive blood culture was obtained (2 per cent). In 92 multiple extractions, positive blood samples occurred in 16 of the cases (17 per cent). In 72 periodontal scalings, positive blood samples resulted in 5 cases (an incidence of 7 per cent). The bacteremias were of very short duration. In the majority of cases, bacteria were detected only within the first minute following completion of the operation. The incidence was greater when moderate or extensive trauma accompanied the operation than when only slight trauma was involved. Significantly greater numbers of bacteremias occurred following extraction of 2 or more teeth than following extraction of single teeth. Variations in tooth mobility and vitality and the presence of periodontal disease or apical infection did not appear to influence the incidence of bacteremia.

17. The Proteolytic Bacteria of the Mouth and the Carious Lesion. *George W. Burnett and Henry W. Scherp, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* Because proteolytic bacteria have been claimed to be the causative agent in dental caries, we have investigated the number, the kind, and the action of proteolytic bacteria found in (1) saliva, (2) enamel caries, and (3) superficial and deep dentin caries. From 144 cases we isolated, on coagulated egg medium, 250 strains of proteolytic organisms consisting mostly of aerobic gram-positive sporulating bacilli and gram-positive cocci, and a few gram-negative bacilli and cocci. A study of the proteolytic activity of these bacteria on gelatin, casein, coagulated blood serum, coagulated egg and decalcified dentin and enamel indicated a close correlation between their abilities to liquefy coagulated egg and to digest decalcified enamel and dentin. No anaerobic bacteria that could digest decalcified enamel and dentin were isolated. Aerobic bacteria that could liquefy decalcified dentin were almost completely absent from the saliva of individuals without open carious lesions, from enamel caries, and deep dentin caries, but were found in small numbers in the saliva of persons having open carious lesions. In the superficial layers of dentin caries, the proteolytic bacteria constituted 7 per cent of the total flora. Intact enamel and dentin were subjected to the action of the liquefying bacteria in broth for periods up to five months with no apparent action. Because of this fact and because such proteolytic bacteria could not be found in the advancing carious lesion, their role as the causative agent of dental caries remains questionable.

18. The Action of Ammonia Salts on Lactobacillus Acidophilus (4646). *Sholom Pearlman, D.D.S., Western Reserve University, Cleveland, Ohio.* Studies have been made of the biochemical mechanisms whereby the growth of *L. acidophilus* (4646) is inhibited in the presence of certain salts of ammonia. Glycolytic activity of nonproliferating cells was measured in the Warburg respirometer by CO_2 evolution from bicarbonate buffer at pH 7.4. Activity was suppressed in the presence of either K^+ or NH^+_4 buffers at that pH. This inhibition was in proportion to the concentration of the added salts, irrespective of the cationic constituent. Growth in an alkaline glucose-beef infusion broth was studied by the dilution-plate method. Addition of K^+, Na^+, or NH^+_4 phosphate buffers depressed the logarithmic growth curves. Inhibition was proportional to the concentration of the added buffers. Similar results were obtained with citrate buffer. No inhibition was noted when NH_4CL or $(NH_4)_2SO_4$ were used, at the same initial pH. In general, inhibitory effects were obtained if the pH of the buffer was between pH 6 and 8. Ammonia salts were not significantly more toxic than salts of potassium or sodium.

19. Serological Relationships Among Certain Lactobacilli Isolated From Foods and From the Oral Cavity. *Frank J. Orland, D.D.S., Ph.D., The Walter G. Zoller Memorial Dental Clinic and the Department of Bacteriology and Parasitology, University of Chicago, Chicago, Ill.* To learn more about the relationship of bacteria of the lactobacillus group isolated from various foods and the oral cavity, a study was made of the antigenic pattern of such strains by means of cell agglutination and agglutinin absorption tests. Major antigens could be demonstrated in certain strains from among the 250 lactobacilli received from other investigators or isolated in the laboratory. These antigens were assigned letter designations following the precedent set by N. B. Williams working with oral strains in this laboratory two years ago. The antigen *F*, for example, was observed in a number of strains, among which, those designated as *L. casei* predominated. This same antigen was also found in a number of strains isolated from the oral cavity of individuals having dental caries. Upon further observation, it was learned this *F* antigen was a rather stable attribute associated with an *L. casei*-type culture possessing the ability to ferment two pentose sugars, rhamnose and sorbose. Other antigens designated *G* and *H* were found in a strain of *L. plantarum* and another *L. casei*, respectively, neither of which exhibited an *F* antigen nor fermented these sugars. The demonstration of such a close relationship between certain strains of oral lactobacilli and strains originally isolated from foods provides pertinent information as to the role of foods in inoculating the oral cavity with these bacteria. Second, there is raised the question of classification of oral bacterial forms shown to be indistinguishable from the L. *casei*-type of microorganism.

20. A Study of the Occurrence of Enterococci, Lactobacilli and Yeasts in Saliva From Man. *Ned B. Williams, Mary Alice Judson, and Charles F. Eickenberg, School of Dentistry, University of Pennsylvania, Philadelphia, Pa.* Interest in the frequency of occurrence of enterococci in the salivas of persons of all ages was stimulated by the isolation and identification of a strain of *Streptococcus faecalis* from a culture obtained from an infected root canal. The salivas were collected in the morning just after arising. The presence of enterococci was indicated by growth and acid production after inoculation and incubation (45 C.) of a broth containing sodium azide, glucose, and bromthymol blue. All positive tubes were studied further for isolation and identification of the microorganisms. Quantitative determinations of lactobacilli and yeasts were made by inoculating tomato juice agar (pH 5.0) with dilutions of the same salivary samples. *Str. faecalis, Str. liquefaciens* or *Str. zymogenes* were identified from 45 of 206 children and adults varying in age from 4 through 40 years. Enterococci were isolated each time from 5 of 10 persons who contributed saliva on from 2 through 7 different occasions. *Str. faecalis* was found sporadically or every time in all salivas while *Str. liquefaciens* was found twice and *Str. zymogenes* once in 3 separate samples from one person. Lactobacilli were found on almost every occasion, but yeasts did not appear with as great regularity. A comparison of the numbers of lactobacilli in the salivas positive for enterococci with those in negative salivas showed that the mean number of lactobacilli in positive salivas was greater than the mean number in negative salivas. A statistical analysis of these data indicated that the difference between these means was significant.

21. Observations on the Chromatin Granules of a Strain of Fusobacterium. *Heiner Hoffman and Matt. C. Dodd, Department of Bacteriology, Ohio State University, Columbus, Ohio.* A positive reaction for nuclear material was

obtained by Robinow's method from the chromatin granules of a strain of fusobacterium. Cytochemical tests for volutin, fat, glycogen, and "iogen" were negative. Examination of wet mounts by phase microscope showed the granules consistently present. The granules evidently go through a developmental cycle associated with cellular division, and filament formation and disintegration. These findings tend to confirm, at least partially, the previous observations of Tunnicliff on the nature of the nuclear-staining granules of fusobacterium.

22. The Proteolytic Activities of Oral Strains of the Genus Candida (Monilia). *Marshall L. Snyder, University of Oregon Dental School, Portland, Ore.* The frequent reversion to alkalinity after initial acidity in saliva-glucose broth (pH 7.4) cultures is nearly always caused by members of the genus Candida (Monilia). This change results from the liberation of basic products from the organic nitrogenous substances in the medium and can be classified under the broad term of proteolysis. Because of recent interest in the proteolytic activity of oral bacteria, the ability of these oral yeastlike forms in attacking various types of protein or protein derivatives and urea was investigated. Strains of Candida were isolated from one hundred individuals whose saliva contained these forms. These were classified culturally: *C. albicans* (82), *C. Kruzei* (8), *C. pseudotropicalis* (1), *C. tropicalis* (3), unidentified (6). These in turn were grown in media containing, as the sole source of organic nitrogen, urea, peptone, and meat extract. In addition, gelatin and coagulated blood serum (Loeffler's medium) were also tested. Only with peptone and meat extract were changes observed indicating release of basic substances. Neither gelatin nor the coagulated blood serum was attacked in 30 days. The results indicated that members of the genus Candida do not attack protein or protein derivatives much higher in structure than peptones, and the latter slowly.

V. Third Session: Saturday Morning, June 25; Abstracts 23-31

23. Studies on Salivary Mucopolysaccharides: Lysozyme-Polysaccharide Complex. *Norman S. Simmons, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* Submaxillary mucopolysaccharide (pig) forms salts with proteins below their isoelectric points. The molecular configuration of these complexes depends upon (1) pH, (2) protein-polysaccharide concentration, and (3) salt. Since lysozyme (egg white) is a basic protein (isoelectric pH 10.5 to 11) the possibility of its forming a complex with submaxillary mucopolysaccharide at the pH level of saliva was investigated. With relatively high concentration of lysozyme a typical mucopolysaccharide-protein clot is formed at low pH levels. At near neutral pH this complex is evidenced by turbidity changes and gelation. The complex is altered at increasing salt levels. With low salt the activity of lysozyme is completely inhibited by the mucopolysaccharide at salivary pH levels. This effect is also diminished with increasing salt concentration.

24. Decalcification Potential of Foodstuffs. *B. G. Bibby and Eugene Chen, Eastman Dental Dispensary, Rochester, N. Y.* A "decalcification potential" derived from the amount of acid formed from foodstuffs and the amount of these foods adhering to the teeth after eating is offered as a tentative basis for comparing the caries-producing capacity of foods. Methods for determining the decalcification potential are outlined and evaluated.

25. Decalcification Indices of Various Foodstuffs. *Eugene Chen and B. G. Bibby, Eastman Dental Dispensary, Rochester, N. Y.* Figures are presented showing the "decalcification potentials" for a number of representatives of various groups of foodstuffs such as raw fruits and vegetables, cooked vegetables or vegetable substitutes, bred and crackers, cereals, cookies and cakes, or candies. Some relationships with temperature of food, fat content, etc., are indicated.

26. Dental Studies in Norwegian Children During and After the Second World War. *Guttorm Toverud, Ph.D., Dental School of Norway, Oslo, Norway.* A special study has been conducted by the pedodontic department of the Dental School of Norway on the influence of wartime on the teeth of children. In collaboration with some school dental clinics in different parts of the country, about 8,000 to 9,000 school children in the age range of 7 to 15 years and 600 preschool children have been studied each year from 1940 to the present time. A preliminary report showing a decrease of about 40 to 50 per cent in D.M.F. teeth and about 60 to 80 per cent in D.M.F. surfaces in deciduous as well as in permanent teeth has been published (Proceedings of The Royal Society of Medicine, London, April, 1949). The present report deals chiefly with the influence of wartime on eruption of permanent teeth during the same period. The different findings in the mouth have been recorded on special charts made up for these studies and later transferred to punch cards. Teeth at least one millimeter exposed in the mouth have been recorded as erupted, or emerged. During the study in 1940-1941 and 1941-1942 all the teeth in the mouth were recorded, but later only the right half of the mouth was taken into account. The children were divided into three groups: city, village, and rural chilrdren as well as into sex and age groups. The last grouping was based upon: (1) the chronological age, and (2) the number of erupted permanent teeth. As previously demonstrated by many investigators, it was found that the girls get their permanent teeth earlier than do the boys. In city boys the time of emergence of the teeth was distinctly delayed from 11 up to 14 years in 1944-1947 compared with 1940-1942. In the girls a similar delay has taken place after the 10th year. In rural children no distinct change in time of tooth emergence could be found. The supposition is made that the delay may partly be explained on the basis of a change in nutrition. Studies in this line are going on. A retardation may also partly be attributed to fewer infected deciduous molars during the latest years with the consequence of longer persistence of them and later emergence of the premolars.

27. The Correlation Between Salivary Lactobacillus Counts and the Rates of Progression of Dental Caries: A Statistical Appraisal. *J. D. Boyd, M.D., Kenneth E. Wessels, D.D.S., and Virgil D. Cheyne, D.D.S., Ph.D., Colleges of Medicine and Dentistry, State University of Iowa, Iowa City, Iowa.* The dental course of 92 teen-aged girls has been followed through frequent oral examinations since July, 1946. In March, 1948, salivary lactobacillus counts were made of each subject, with repeat analyses on the following day. Between March and December, from 2 to 6 additional lactobacillus counts were made for each child. The counts were made in the state hygienic laboratory as a part of the service provided for dentists of the state. Samples were collected in the early morning before mouth cleansing or the eating of food; the usual technic of collection after chewing paraffin was used. The lactobacillus counts ranged from 300 to 960,000, with an average of 115,000. The median value was 81,000. Half of the samples ranged from 50,000 to 135,000. Values for successive days for samples from the individual child generally were in

good agreement. Caries progression from July, 1946, to December, 1948, expressed in terms of newly carious D.M.F. surfaces per annum, ranged from zero to 16.3. The mean value was 3.31 ± 0.34; the median value was 2.2. The median lactobacillus counts from grouped data from subjects with extreme variation in caries progression rates are listed in Table I. The correlation coefficient for the averages of all lactobacillus counts for each subject in terms of their value for caries increment (new D.M.F. tooth surfaces per annum) was 0.1959. This value is not statistically significant. The reliability of the lactobacillus count as a diagnostic or prognostic agent was discussed.

TABLE I

RATE OF ADVANCE OF DENTAL CARIES (NEW D.M.F. SURFACES PER ANNUM)	NUMBER OF SUBJECTS	MEDIAN LACTOBACILLUS COUNT: FROM AVERAGE OF INDIVIDUAL VALUES (MARCH)	MEDIAN LACTOBACILLUS COUNT: FROM AVERAGE OF ALL COUNTS FOR EACH INDIVIDUAL (MARCH THROUGH NOVEMBER)
Those with no new decay or measurable advance	10	60,000	45,000
Those with less than 0.8 new D.M.F. surfaces per annum	17	60,000	50,000
Those with most rapid advance of caries (5.8 or more new D.M.F. surfaces per annum)	9	70,000	70,000

23. THE THERMAL CONDUCTIVITY OF DENTIN. *Vincent F. Lisanti, Tufts College Dental School, Department of Oral Pediatrics, Boston, Mass.* An apparatus has been constructed for the determination of the thermal conductivity of sections of nonvital dentin. The design of the apparatus is based on the principle that heat conducted through a substance of known conductivity will be altered by the insertion of a substance of unknown conductivity. Thermal conductivity is calculated from the mean value of the amount of heat (Q) delivered to and removed from the unknown substance by the known conductor, when "Q" is applied to the formula, $Q = \frac{K \Delta T A}{L}$. "K" is conductivity; "$\Delta$ T" is the difference in temperature (° C.) of the two surfaces of the unknown; "A" is the area of the specimen and "L" is the length of the specimen, $K = \frac{QL}{\Delta TA}$. The apparatus was standardized against a sample of Pyrex glass of known thermal conductivity. The thermal conductivity of polished dehydrated sections of dentin from extracted teeth was calculated for temperatures of 30, 50, and 100° Centigrade. Dentin from unerupted teeth, from teeth of subjects of various ages, and regular and irregular dentin were used in this experiment.

29. STUDIES OF THE ACTION OF SODIUM FLUORIDE ON ENAMEL BY ELECTRON MICROSCOPY AND ELECTRON DIFFRACTION. *David B. Scott, Robert G. Picard, and Ralph W. G. Wyckoff, National Institutes of Health and R. C. A. Victor Division, Bethesda, Md.* Over 500 slabs of enamel, cut parallel to the tooth surfaces, have been immersed in NaF solutions for periods of time ranging from 4 minutes to 30 days. Fluoride solutions were varied in concentration from 2 to 4 per cent, in pH from 4.5 to 8.0; all solutions were filtered, and all containers were paraffin-lined. Control slabs were immersed in water and equimolar NaCl solutions for equal periods of time. Washing after treatment varied from one minute to 6 days in circulating distilled water. After washing, chromium-shadowed collodion replicas of the slabs were prepared and

studied under the electron microscope. Thus far over 1,000 screens have been examined, and 2,600 micrographs taken. A recognizable inhomogeneous apparently crystalline surface deposit was observed after 15 to 30 days' immersion in all fluoride solutions. From 40 hours' to 7 days' immersion resulted in traces of deposit, and under 40 hours, results were largely negative. The deposit after 30 days' immersion was still apparent after 24 or 72 hours' washing, but not after washing for 6 days in circulating water. Electron diffraction of the surfaces of 75 slabs before and after similar treatment with NaF has indicated that the deposit formed is CaF_2. A much slighter deposit of CaF_2 can be detected by electron diffraction than by electron microscopy. After only 3 minutes' treatment with 2 per cent NaF, the normal apatite pattern has been replaced by the typical pattern of CaF_2. The same pattern has been seen after all longer treatments with NaF and in all experiments involving more concentrated solutions. Preliminary washing experiments after 3-minute exposures have shown a reversion to the original apatite pattern after 90 minutes' washing. Experiments indicate that more prolonged washing is needed to re-establish the apatite pattern after longer treatments with NaF. Studies are in progress to determine further the significance and permanence of these surface films of CaF_2.

30. The Architectural Pattern of the Epithelio-Connective Tissue Junction of the Gingiva in the Rhesus Monkey. *R. D. Emslie, I. Schour, and J. P. Weinmann, School of Dentistry, University of Illinois, Chicago, Ill.* The study was based on the examination of 60 gingival biopsy specimens taken from 14 Rhesus monkeys. Separation of the epithelium from the lamina propria was achieved after maceration of the specimens in 0.5 per cent acetic acid for 6 hours. The architectural pattern was studied with a dissecting microscope. The alveolar mucosa (lining mucosa of the alveolar process) showed the fundamental pattern of the epithelio-connective tissue junction. The epithelium was not characterized by so-called epithelial pegs, but formed a flat sheet, pitted at intervals for connective tissue papillae. The masticatory mucosa (free and attached gingiva and palatal mucosa) was characterized by an increase in the number and size of the connective tissue papillae, providing increased nutrition for the epithelium. This produced a honeycomb appearance in the epithelium, with the ridges in one direction usually accentuated. Contrary to current teaching, the epithelial attachment was also pitted for connective tissue papillae. The latter ran parallel to the long axis of the tooth. In mild chronic inflammation the design of the epithelial honeycomb was not changed, but the epithelial ridges were thickened. An increase in the severity of the inflammation produced a downgrowth of some of the epithelial ridges, forming a much coarser honeycomb with large "cells" pitted at their blind ends for connective tissue papillae. The routine use of a separation technic as an adjunct to the usual histologic methods in studying human gingival biopsy specimens may have diagnostic value.

31. A New Machine for Grinding Tooth and Bone Sections. *Peter P. Laude, D.D.S., College of Dentistry, State University of Iowa, Iowa City, Iowa.* This machine is designed for grinding tooth and bone sections easily and quickly to a thickness of less than three thousandths of an inch. The grinding disk is regulated by a control knob which is calibrated into fifty spaces of one thousandth of an inch each. If the section is measured by a micrometer before mounting on the machine, its thickness can be determined at any time during the grinding by noting the number of spaces through which the control knob has been revolved after the grinding disk makes contact with the section.

VI. Fourth Session: Saturday Afternoon, June 25; Abstracts 32-43

32. Solubility of Enamel, Fluorapatite, Calcium Fluoride, and Tricalcium Phosphate. *Thomas M. Boyd, D.D.S., Indiana University School of Dentistry, Indianapolis, Ind.* Gerould compared in a rough manner the solubility of calcium-hydroxyapatite, calcium fluorapatite, and fluorite using N/1 HCl. This study is a more careful examination of the solubility of these three substances compared with powdered enamel. One hundred milligram samples were decalcified for 40 minutes in 25 ml. of a standard sodium acetate buffer. All four substances were screened through a 325-mesh sieve to insure uniform particle size, and each substance was subjected to two different buffer solutions, pH 2.5 and 4.0, respectively. The solubility of each substance was examined by both weight loss of sample, and calcium analysis on the acid filtrate. An average of ten determinations on each substance indicates that calcium fluoride is the most insoluble of the four substances tested, whereas calcium fluorapatite, powdered enamel, and tricalcium phosphate are the next least soluble in that order. All samples experienced a greater weight loss and more calcium was detected in the filtrates following decalcification in the pH 2.5 buffer. The data demonstrated that either of the supposed end products of a reaction between a soluble fluoride and enamel, that is fluorapatite or calcium fluoride, is definitely less soluble in weak organic acids than would be the unprotected enamel surface.

33. The Determination of Amino Acid Content of Fresh and Putrefied Saliva by Paper Partition Chromatography. *L. S. Fosdick and R. Q. Blackwell, Northwestern University School of Dentistry, Chicago, Ill.* The amino acid content of caries-active, caries-immune, and periodontal disease saliva was determined on saliva samples, both fresh and following varying lengths of incubation. One-dimensional paper chromatography was used for the determinations. Six caries-active, four caries-immune, and three periodontal disease cases were studied. On the basis of the limited number of cases studied, it would appear that higher concentrations of aspartic and glutamic acids occur in caries-active saliva than in periodontal disease saliva with intermediate amounts present in caries-immune saliva. More work, which is being carried on in this laboratory at the present time, is necessary to establish the statistical significance of the apparent trend.

34. Some Amino Acid Derivatives of p-Hydroxybenzoic Acid. *E. B. Tooper and L. S. Fosdick, Northwestern University School of Dentistry, Chicago, Ill.* It has been known for some time that hydroxybenzoic acids have a pronounced antiseptic action (Kolbe, 1874). For this reason it was believed that certain derivatives of p-hydroxybenzoic acid, designed to be retained by the mucin plaque, might prove to be enzyme inhibitors. It seemed probable that a peptide-like substance would be best retained in the mucin. p-hydroxybenzoic acid was coupled to the amino acids, glycine, alanine, phenylalanine, aspartic acid, glutamic acid, methionine, and leucine, and tested for inhibition properties. In the concentrations used, all either showed no inhibiting properties or were very mild in their action.

35. The Synthesis and Enzyme Inhibitory Activity of Some Amino Acid Derivatives of Sulfanilamide. *L. S. Fosdick and O. Norman, Northwestern University, School of Dentistry, Chicago, Ill.* On the basis that certain peptides of the antibiotic compounds will retain their enzyme inhibitory activity in the mouth for prolonged periods of time, it was thought that if amino acids or amino acid derivatives could be found which exhibited enzyme inhibitory

activity, that they also may remain in the mouth and exert a prolonged influence. For this reason amino acid derivatives of p-amino sulfonic acid were prepared. All of the compounds were prepared by reacting an appropriate amino acid with benzene sulfonil chloride. In cases where a dipeptide chain was introduced, the acid chloride of the amino acid derivative was prepared by the use of phosphorus beta chloride, after which the acid chloride was reacted with the appropriate amino acid. The amino acids used in this study are glycine, dl-alanine, dl-phenylalanine, dl-valine, dl-norvaline, dl-leucine, l-leucine, dl-isoleucine, and dl-norleucine. None of the derivatives prepared today exhibit the enzyme activity of the antibiotics.

36. The Synthesis of Inhibitory Activity of Compounds of the Quinone Type. *J. C. Calandra and E. Adams, Northwestern University School of Dentistry, Chicago, Ill.* A series of amino derivatives of 2,3-dichloro-1,4-naphthoquinone were prepared and their activity as enzyme inhibitors of the degradation of carbohydrate was determined. The compounds synthesized were the sulfonamide, amino acid, alkyl amine, amino acid, amino pyridine, and guanidine derivatives of the quinones. They were prepared by heating the amino compounds with the quinone in alcohol for sixteen to twenty hours, after which the reaction mixture was allowed to cool and the products were isolated in the usual manner. In some cases a trace of diethyl amine was added as catalyst. In cases where the derivative was insoluble they were used as the sodium salts. All of the water-soluble derivatives exhibited enzyme inhibitory activity with the exception of the sulfonamide derivatives, which were less effective. The guanidine carbonate derivative was especially effective.

37. Studies of the Bacterial Plaque and Dental Caries. II. Method for the Isolation of Actinomyces Isreali From Bacterial Plaque. *J. Ennever, College of Dentistry, Ohio State University, Columbus, Ohio.* The bacterial plaque is the nidus for the production of organic acids associated with the initiation of dental caries. This laboratory has been concerned with determining the role played by *Actinomyces isreali* in the plaque structure. However, the methods for isolation of the parasitic actinomycetes from contaminated sources have been inadequate, primarily because of the difficulty in securing a uniform inoculum. The essentials of the method employed are: (1) preparation of a uniform suspension of plaque material, (2) transfer of visible granules to brain heart infusion agar, and (3) anaerobic incubation at 37° C. with 5 per cent CO_2 for three days. Utilizing this procedure, consistent recovery of *A. isreali* from a bacterial plaque was demonstrated.

38. A Biologically Active Salivary Fraction Possibly Related to Caries Susceptibility. *T. J. Hill, B. White, M. Matt, and S. Pearlman, Western Reserve University, Cleveland, Ohio.* Saliva has been fractionated and a portion obtained which may be an influence in dental caries susceptibility. The extract has been recovered in quantities of from .01 to .04 mg. per ml. of saliva. In these preliminary extractions, larger amounts have been recovered from salivas of people resistant to dental caries. This extract inhibits the growth of certain bacteria; when injected into animals, it produces a prolonged hyperglycemia which suggests that its action may be the interference with carbohydrate metabolism. When added to whole fresh saliva, it retards the rate of acid production. When incorporated in dextrose broth, the rate of growth of *Lactobacillus acidophilus* is in inverse relation to the amount added. Two milligrams per milliliter completely stops the growth of lactobacilli. Smaller amounts suppress the growth for periods of 24 to 48 hours. Injection of salivary

extract into rats increased the peak of the glucose tolerance curve above that which could be explained upon the basis of fright reaction to the injection. The return of the blood sugar level to normal was delayed. The nature of the material has not yet been determined. It is known to be soluble in water but insoluble in alcohol or ether. It is thermostabile and not dializable. Efforts to identify the material are now being made.

39. Salivary Ammonia and Its Correlation to Dental Calculus. *Marvin Jacobson, D.D.S., M.S., and Robert G. Kesel, D.D.S., M.S., University of Illinois College of Dentistry, Chicago, Ill.* In investigating a possible mechanism of dental calculus formation, the ammonia nitrogen developed from the supernatant liquid and the residue of centrifuged stagnating saliva over a nine-day incubation period was studied. One hundred two patients were used in this study. They were classified as to calculus present, calculus free, and periodontal diseased. Approximately 5 c.c. of paraffin-stimulated saliva was first obtained after which another 50 c.c. was received in another sterile container. The first sample was used for the *Lactobacillus acidophilus* determination. The second sample was used for the ammonia determinations on the initial day and on the third, fifth, seventh, and ninth subsequent days. The residue was separated from the supernatant liquid by centrifugation at 3500 r.p.m. for twenty minutes. One cubic centimeter of the supernatant liquid was used for the determination and the drained sediment for the residue determination. The results show that the greatest concentration of ammonia nitrogen was found in the supernatant liquid. The periodontal diseased group had the greatest concentration, the calculus free and the calculus present about the same concentration. It was shown that bacterial action was necessary for ammonia production. Statistically there was no difference in the ammonia production of the calculus-free group compared to that of the calculus-present group.

40. The Softening and Solution of the Dentin in Caries. *Milton B. Engel, University of Illinois, College of Dentistry, Chicago, Ill.* The ground substance of dentin consists largely of a carbohydrate-protein complex which is believed to be highly polymerized. Unaltered normal adult dentin does not give a reaction for polysaccharides when stained by the Hotchkiss periodic leucofushsin method. Carious dentin, however, stains readily. This is due to the unmasking of reactive groups and is presumed to be evidence of depolymerization. When dentin which has been initially treated with N hydrochloric acid is incubated with an enzyme collagenase, the dentin becomes soft and ultimately dissolves. These changes may be observed by physical method and by assay of collagenase-dentin filtrates for soluble nitrogen. Dentin treated in this way shows many of the characteristics of carious dentin when stained by the Hotchkiss procedure. Water extracts of lyophilized carious dentin which are assayed for enzyme activity show the presence of two factors. One factor gives reactions characteristic of a "collagenase"; the other activity is characteristic of a mucolytic enzyme of the hyaluronidase group. It is concluded that the softening and solution of the dentin in caries are in part the result of the action of a collagenase. The hyaluronidase is believed to be active in causing the pulpal reaction in advanced caries.

41. The Relative Occurrence of Pits in Human Deciduous Molars and the Possible Relationship to Nutrition. *Gerald J. Cox, Ph.D., and C. W. Hagan, D.D.S., School of Dentistry, University of Pittsburgh, Pittsburgh, Pa.* In 296 pairs of upper and 202 pairs of lower deciduous human molars from one side of the mouth, pits were observed in the percentages shown in tabular form. In 176 upper second molars the supplementary cusp was observed with a fissure at the junction with the lingual surface occurring in 49.4 per cent of the cases.

TABLE I

SURFACES	UPPER MOLARS		LOWER MOLARS	
	FIRST (%)	SECOND (%)	FIRST (%)	SECOND (%)
Buccal		2.7		
Occlusal, mesial	6.8	18.2	5.4	24.2
Occlusal, central	3.7	23.0	10.4	33.7
Occlusal, distal	15.2	54.7	20.3	35.7
Lingual		14.5		11.4
Mesiobuccal				10.4
Distobuccal				5.9

Pits were found more frequently in the occlusal surface of the second than of the first molar. The findings of Cox and Levin (*J. D. Res.* **23**: 191, 1944) suggest that comparably better nutrition during the formation of first molars results in fewer pits in these teeth than in second molars.

42. MEASUREMENT OF MASTICATORY PERFORMANCE AND EFFICIENCY. *R. S. Manly, Ph.D., and Louise C. Braley, B.S., Tufts College Dental School, Boston, Mass.* The particle size distribution of masticated peanuts was studied by sieving on screens ranging from 10 to 200 mesh. Tests were made on 10 subjects at 5, 10, 20, 40, 80, and 160 chews. Food chewed only 5 times had a size distribution characteristic of random grinding processes, for the screen size data gave a straight line when plotted against the percentage oversize on logarithmic probability paper. Peanuts chewed more than 5 times gave distribution lines which became progressively more curved in a direction which indicated an abnormally rapid disappearance of larger particles. This finding suggested that mastication tends to reduce the percentage of larger particles rather than to grind all sizes uniformly and was adopted as a test for masticatory performance. The precision of the test was satisfactory since the self-correlation coefficient of 25 repeated observations was 0.91. Variation in weight of individual portions from 3 to 7 grams did not affect performance providing the number of chews applied to the whole sample remained the same. The percentage of food passing a 10-mesh screen increased uniformly but not linearly with increasing number of chews. It was proportional to the logarithm of the number of chews within limits. The average masticatory performance of one side of a "normal" dentition (with or without third molars) was 83 per cent (46) through 10 mesh. With first or second molar missing, it was 57 per cent (17), and for denture wearers, 35 per cent (31). On the basis of certain assumptions the masticatory efficiencies of the three groups were 100, 36, and 18 per cent, respectively.

43. CITRATE CONTENT OF SOUND AND CARIOUS HUMAN TEETH. *Isadore Zipkin and K. A. Piez, National Institute of Dental Research, National Institutes of Health, Bethesda, Md.* The crowns of whole human teeth were separated into dentin and enamel by the Manly-Hodge technic, and analyzed for citric acid by a modification of the Perlman, Lardy, and Johnson method. Sixty-five samples of sound dentin, 129 samples of carious dentin, 39 samples of sound enamel, and 83 samples of carious enamel comprised the study. The analyses were repeated after approximately one year to determine the stability of citrate in dentin and enamel. No significant change occurred after one year's standing in either the dentin or enamel. The dentin of sound and carious teeth contained 928 mg. per cent and 884 mg. per cent citric acid, respectively, and the difference between the means was not significant. The enamel of sound and carious teeth contained 100.9 mg. per cent and 97.4 mg. per cent citric acid, respectively, and the difference in the means was not significant. It was also shown that there was no significant variation in the citrate content of teeth according to their morphology.

VII. Fifth Session: Saturday Evening, June 25; Abstracts 44-55

44. Dental Diseases and Nutrition in Puerto Rican Children. *K. L. Shourie, J. W. Hein, S. W. Leung, N. S. Simmons, and C. D. Marshall-Day, School of Medicine and Dentistry, University of Rochester, and Eastman Dental Dispensary, Rochester, N. Y.* Supplementing a dental survey of Puerto Rican children, data were collected regarding the incidence of deficiency diseases, height and weight, economic status, and availability of school lunches. Information about fluoride content of drinking water and length of residence was collected. Quantitative dietary studies of 15 representative families in each of six areas visited were carried out. Of 1,754 children examined for clinical signs of deficiency diseases, only 0.7 per cent were classed as deficient. For 622 boys, 14 years old, given a detailed clinical examination, caries incidence increased as weight increased (7.5 D.M.F. teeth per child in lowest weight group and 9.5 in the highest weight group). Children consuming school lunches showed a lower average D.M.F. Economic status did not appear involved. Fluoride content of drinking water in Puerto Rico usually is either negative or 0.1 p.p.m. However, the town of Carolina, where children have the lowest D.M.F. has 0.4 p.p.m. of fluoride. The length of residence in the metropolitan area may be a factor in the caries incidence and is shown by the fact that children living in San Juan from birth showed an average of 10.3 D.M.F. teeth, as against 9.2 for those living there for 8 years or more, and 8.8 for those of less than 8 years' residence. The variations in the caloric values and quantities of principal dietary constituents between the six localities were not reflected in variations in caries or gingivitis incidence.

45. Dental Caries Studies in Puerto Rican Children. *S. W. Leung, K. L. Shourie, J. W. Hein, N. S. Simmons, and C. D. Marshall-Day, School of Medicine and Dentistry, University of Rochester, and Eastman Dental Dispensary, Rochester, N. Y.* Examinations for the number of decayed, missing, and filled teeth in 2,099 male and female Puerto Rican children between the ages of 6 and 18 years gave an over-all group average D.M.F. rate of 7.3 Boys showed a somewhat higher average score than girls, the values being 7.6 and 6.9, respectively. A breakdown by age groups indicates, however, that higher rates for males existed only in the age range 6 to 12 years. At age 14 the score was similar for both sexes, while for ages 16 to 18 the girls showed a slightly higher incidence of caries attack. A more detailed examination was done on additional group of 622 14-year-old boys. The clinical examinations included the charting of the location and relative size of each lesion and were supplemented by full-mouth bite-wing radiographs. The mean D.M.F. for this group was 8.4, representing an increase of 1.2 over that of the 14-year-old boys in the first study. The increase may be largely attributed to the use of roentgenograms. Further analyses showed that there was an average of 4.8 occlusal cavities per subject as against 6.2 interproximal cavities. A larger percentage of interproximal cavities was found on those surfaces in which mesial-distal contacts were present. On an average there was the loss of 0.7 teeth per subject from extractions, the lower first molars being most frequently involved. The average number of fillings per child was only 0.78.

46. Oral Bacteriological Studies in Puerto Rican Children. *C. D. Marshall-Day, K. L. Shourie, J. W. Hein, S. W. Leung, and N. S. Simmons, School of Medicine and Dentistry, University of Rochester, and Eastman Dental Dispensary, Rochester, N. Y.* Samples of saliva collected from 564 14-year-old boys from six Puerto Rican localities were air mailed to the bacteriological

laboratory of the New York State Department of Health for *Lactobacillus acidophilus*, cocci, and yeast counts. In 312 of these cases duplicate samples were sent to the Michigan Caries Control Laboratory for lactobacillus counts only. The average period between collection and analyses was 3 days, the maximum being 4 days. *L. acidophilus* counts of the duplicate samples showed a high coefficient of correlation between the two laboratories (0.74). The standard product moment correlation between the lactobacillus counts and the D.M.F. teeth, number of open cavities (excluding roots), and D.M.F. areas were only 0.30, 0.33 and 0.33, respectively. Similar correlation between cocci and D.M.F. teeth gave a coefficient of 0.27. The correlation coefficients between lactobacillus counts and the amounts of gingival involvement and the amount of debris around the teeth were much lower. The data show no evidence that the l.a. count is closely correlated with either the cumulative caries experience or the gingival condition of the children.

47. Gingival Disease in Puerto Rican Children. *J. W. Hein, K. L. Shourie, S. W. Leung, N. S. Simmons, and C. D. Marshall-Day, School of Medicine and Dentistry, University of Rochester, and Eastman Dental Dispensary, Rochester, N. Y.* A total of 2,265 Puerto Rican children, 6 to 18 years old, were examined for gingival disease. In 1,646 children the incidence and degree of gingivitis were recorded. Both sexes showed a higher incidence of gingivitis at the age of 12 years, although girls averaged for all age groups less than boys. Six hundred twenty-two boys, 14 years old, were given periodontal examination which included observation of each labial or buccal margin and papilla for presence of hyperemia, proliferation, and edema. Analysis of data showed that proliferation and edema tend to be generalized. The upper posterior gingivae were the most affected, the lower posterior gingivae least affected. Adjacent margins and papillae were simultaneously affected by either hyperemia, proliferation, or edema. The incidence of hyperemia, proliferation, and edema in the upper anterior margins or papillae reflected to some extent a similar state for the whole mouth. A general impression of the gingival conditions in the anterior region of the mouth was found to be 90 per cent accurate in reflecting clinically normal mouths but not when estimates regarding incidence of gingival disease were involved. Measurements of pocket depth in the region of the buccal margin of the lower left first incisor and the upper right first incisor indicate that as pocket depth increases the percentage of cases with proliferation decreases, while the number of cases of hyperemia and edema increase. No definite difference between the caries incidence in subjects with clinically normal gingivae and total group was found.

48. Subcrevicular Detachment Preceding Periodontal Pocket Extension. *R. Gordon Agnew, B.A., M.Sc. (Dent), L.D.S., D.D.S., Ph.D., University of California, San Francisco, Calif.* Various concepts of pocket formation, while differing materially in underlying principles and in sequence of tissue changes, have assumed a more or less even and progressive stripping away of epithelium from tooth surface at the pocket or crevice base. Where the cuticula dentis has formed at the time of such separation, this hornified membrane is commonly left adherent to the hard tissue surface. Recent studies have amplified observations first published in 1933 indicating that, under certain conditions, single or multiple areas, varying in size from circa fifty to several hundred micra, may be found between epithelial attachment and root surface at varying distances below the pocket base in which the epithelium has already separated from the tooth surface. The hornified secondary cuticle (cuticula dentis), if formed at that time, is almost invariably located on the soft tissue

side, in contradistinction to the classic pattern in pocket formation. Serial section study reveals that these zones resemble isolated bubbles on the cementum, or may extend in an irregular fashion along the root surface. In single sections they show a semilunar or hemispherical contour. Confusion with artefact is usually readily eliminated. As these structures are approached by the deepening crevice the friable cuticular rim is commonly fractured and the crevice, at that point, deepens abruptly. Such scalloped contours formed by the adherent cuticle on the soft tissue wall may persist, ultimately being located well above the crevice base, and may be filled with calcular deposit indicating their existence in vivo. Where the cuticular rim has not fractured the epithelium may peel away from the cuticle, ultimately leaving bubble-like structures on the root surface at varying distances from the crevice base. Calculus may, on occasion, be seen in a subcrevicularly located detachment; in such an instance serial study will reveal contact with the surface at some near-by point. These detachments may occur not only along the marginal epithelial attachment but also in root bifurcations and trifurcations, and along the apical surface in connection with epitheliated granulomata. This detachment, for which the term "subcrevicular detachment" is proposed, suggests a sporadic conditioning or biologic alteration of the tooth surface in advance of the usual denudation process. Marked gingival inflammation is a common accompaniment and may be causally related. However such inflammatory states are seen so commonly in the absence of this phenomenon that it might be postulated that local and general factors combine in its etiology. The zones are cystlike in appearance, although no cystic contents have yet been identified. Nutritional influences might affect the areas in question from the subepithelial tissue of the immediate neighborhood or, hypothetically, and to a questionable degree, from a vital pulp through the cementum. The condition has been seldom observed in experimental animals although recently studied in a Java monkey. In the human being it has been seen with relative frequency in systematic autopsy study. No single age bracket or systemic state has been uniformly associated with its occurrence. Generalized arteriosclerosis has been commonly noted in older individuals. These zones, especially when large or multiple, will obviously lead to abrupt pocket extensions; they may, in part, account for incompleteness of the cuticular coat residually left on the cementum, or for those instances where calculus, in discrete areas, strips readily from exposed cementum.

48. JUVENILE PARADENTITIS. *O. J. Mikola and Wm. H. Bauer, St. Louis University School of Dentistry, St. Louis, Mo.* The microscopic findings of jaws removed at autopsy from four children aged 10, 14, 17, and 18, who succumbed to systemic diseases, are reported and compared with similar changes in jaws of adults. In all cases inflammatory processes of the subepithelium of interdental papillae were seen to proceed apically penetrating and loosening the transseptal fibers of the periodontal membrane. The influx of inflammatory cells into the alveolar crest region stimulates the differentiation of osteoclasts resulting in a resorption of the crest and in many instances perforation through the crest into the underlying bone marrow spaces. It is the advancing inflammation which separates the Sharpey's bundles from the cementum and alveolar plate and causes the cells of the epithelial attachment to proliferate along the cementum. The apical progression of the inflammatory process stimulates the downward fingerlike proliferation of the surface epithelium and epithelial attachment. Although severe juvenile paradentitis was observed in other children free of systemic disease (not reported herein), it is likely that various systemic involvements, as a predisposing factor for paradentitis, play a more important role in juveniles than in adults. Accordingly diseases of long standing render

the tooth-supporting structures more susceptible and less resistant to inflammatory processes. The rapidity of the advance of the inflammation along the periodontal membrane is enhanced by the altered function which is due to an unfavorable change of balance between the clinical crown and invested roots.

49. The Gingiva in Leukemia. *F. M. Wentz, G. J. Anday, and B. Orban, School of Dentistry, University of Illinois, and Loyola University School of Medicine, Chicago, Ill.* A statistical evaluation of oral symptoms was made on the basis of 149 case histories. The clinical symptoms consisted of ulceration, pallor, hemorrhage, and hypertrophy of the gingiva. The clinical symptoms were more severe in acute than in chronic leukemia. Biopsy specimens were obtained from 13 patients suffering from acute or chronic myeloid or lymphatic leukemia. The acute types showed histologically specific gingival changes. These consisted of dense infiltration of the lamina propria by white blood cell elements of immature types. Mitoses of these forms was frequently noted. An oral smear in a case of acute myelogenous leukemia demonstrated the immature white blood cell elements in a gingival pocket. No specific histologic changes were observed in the chronic forms of leukemias. Other tissue changes of the gingiva seemed to be incidental and were apparently not related to the systemic condition.

50. Biomicroscopic and Histologic Evaluation of Normal and Inflamed Gingivae in the Albino Rat. *Irving Glickman and Leif Johannessen, Tufts College Dental School, Boston, Mass.* In 150 albino rats, a correlated, gross, biomicroscopic, and histologic study was conducted dealing with the gingival capillary response to chemical irritation and the evaluation of the reliability of the biomicroscope as an indicator of gingival changes. A single application of a 6 per cent solution of freshly prepared chromic acid was used to create gingival inflammation. Correlation of the findings indicated the following: In normal gingiva, the biomicroscope revealed a regular capillary pattern and corpuscle flow not detectable by gross examination. A single application of chromic acid resulted in initial erythema, followed in two hours by gingival degeneration and necrosis. Gingival repair with an attempt at restoration of gingival contour occurred within the eight-day experimental period. The biomicroscope was more effective than gross examination in the delineation of early gingival circulatory changes in response to local irritation, but did not contribute particularly to the evaluation of necrotic changes.

51. Periodontal Disease in the Hamster: Dietary Consistency, Coprophagy, and Maintenance Factors. *David F. Mitchell, University of Rochester, School of Medicine and Dentistry, Rochester, N. Y., and University of Minnesota, School of Dentistry, Minneapolis, Minn.* In the past, periodontal disease has been produced in hamsters maintained under the following conditions: (1) fed high carbohydrate fine powder diet (CPD), (2) kept in cages with wide mesh wire bottoms, allowing most of feces to fall from reach, and (3) without wood shavings for bedding. By controlling maintenance conditions, these three principal variables were tested in order to determine optimal conditions for the production of periodontal disease. Seventy-five animals, 32 days old, were distributed according to littermates and sex into 5 groups. After 98 days the animals were sacrificed. A method was devised by which the jaws were scored for periodontal disease. Practically all the animals exhibited some degree of this disease. The *type* of disease varied with coarse (laboratory chow) and fine (CPD) diet and with presence or absence of shavings. The *amount or degree* of affliction, as measured, was much greater with the CPD than with the

chow. Partial restriction of coprophagy had no effect on type or amount of the disease. Shavings were impacted subgingivally in some jaws, but the over-all incidence did not vary with this factor. No sex difference in the incidence of disease was seen. The incidence of supragingival caries varied only with the type of diet—high: CPD. Microscopic studies of palatal, gingival, and esophageal keratinization of epithelium showed no variation with these factors. Dental caries occurred in intimate association with periodontal lesions.

52. Fusion Frequency as an Indication of Pulp Condition. *D. Y. Burrill, University of Louisville, School of Dentistry, Louisville, Ky.* Using a relaxation oscillator delivering peaks of current lasting approximately 0.0013 second, at a frequency variable from 2 to 1160 per second, with a maximum current delivery of about 40 milliamperes, tests were made to determine the lowest frequencies that would produce continuous rather than vibratory or beating sensations in the pulps of teeth, the fusion frequency. Seventy-two normal teeth were found to give a fusion frequency of 9 or more per second, with a range of 9 to 24, a median of 13, and an average of 13.76. Nineteen teeth considered questionable for various reasons, but without pulp exposure, had fusion frequencies ranging from 3 to 13 per second with a median of 9 and an average of 8.4 per second. Twenty-five teeth with carious pulp exposures but no definite evidence of pulp pathosis had fusion frequencies ranging from 3 to 13 per second, with a median of 4.2, and an average of 4.6 Ten teeth with carious pulp exposures and definite clinical evidence of pulp pathology had fusion frequencies ranging from 2 to 6 per second, a median of 3, and an average of 3.6 per second. Five known nonvital teeth gave fusion frequencies above 24 per second. At least one normal tooth was tested at the same time as each known or suspected diseased tooth. These figures suggest a possible test for pulp condition if further studies confirm the above results.

53. In Vitro Studies of the Effects of Some Chemical Substances on the Growth of Oral Microorganisms and on Their Ability to Dissolve Tooth Salts. *Robert M. Stephan, National Institute of Dental Research, National Institutes of Health, Bethesda, Md.* Using an "auxanographic" culture plate method, a comparison has been made of the stimulatory and inhibitory effects of salts of various elements on the growth of oral lactobacilli, streptococci, staphylococci, and a few other microorganisms. The media employed were of the chemically defined type, except for the use of an acid hydrolysate of purified casein, the use of a specially purified agar, the omission of the salts of trace elements, and the addition of $CaCl_2$ and Na_3PO_4 to form a calcified precipitate. The initial pH before incubation was around 7.0. The concentration of salts applied to culture plates with filter paper discs, or agar plugs was 0.01 molar for Mn^{++}, Fe^{+++}, Co^{++}, Cu^{++}, Zn^{++}, F^{-}, Mg^{++}, and Ni^{++}; 0.1 molar for K^{+}, while 5 per cent concentrations of Na_2HPO_4 and $(NH_4)_2HPO_4$ were employed. Manganese stimulated or was essential for the growth of 28 of 30 strains of lactobacilli, but was inhibitory to staphylococci and streptococci. Iron and zinc showed both stimulatory and inhibitory effects, whereas cobalt, copper, nickel, molybdenum, and fluorine showed only inhibitory effects for all organisms tested. Potassium was required or was greatly stimulatory to most of the microorganisms tested, and magnesium was greatly stimulatory to only a few. With the lactobacilli and to a lesser extent with the streptococci, dissolution of the calcium phosphate precipitate followed growth of the colonies, and was accompanied with a lowering of pH produced in the medium around the colonies. Only slight stimulatory or inhibitory effects were observed with either Na_2HPO_4 or $(NH_4)_2HPO_4$ for the 30 strains of lactobacilli studied, and no significant difference was observed in the effects produced by the sodium and ammonium ions.

54. The Effects of Alloxan Diabetes on the Incidence of Dental Caries in the Syrian Hamster. *Frank G. Everett, Daman Lal, and Nilkanth M. Phatak, University of Oregon Dental School and University of Oregon Medical School, Portland, Ore.* Four groups of 35 hamsters each, fifteen days after weaning, were set up in this investigation. Two groups were made diabetic (to the point of producing definite glycosuria) by administration of alloxan monohydrate. One of these two groups was placed on a caries-producing high sugar diet, the other on a noncariogenic maintenance diet, both diets ground to pass mesh No. 40. The remaining two groups served as controls and consisted of normal animals on the respective type diets mentioned above. The experiment was continued for 135 to 145 days, when the animals were sacrificed and their dental caries extent was scored following Keyes' method. The results to date indicate that surviving diabetic animals (9) on cariogenic high sugar diet had a caries experience approximately double that of its control group of normal animals (29) on the same diet, and approximately 10 times greater than that of equally diabetic surviving animals (4) but on a noncariogenic diet. However, even on this noncariogenic diet the diabetic animals (4) produced a small but appreciable caries score compared to its own healthy controls (30 animals) on the same diet, producing none. Animals (52) recovering from alloxan effects showed intermediate caries scores.

55. Dental Caries in the Syrian Hamster. VI. The Effect of Gonadectomy and Testosterone Propionate on Caries Activity. *Paul H. Keyes, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* In previous studies male hamsters were more susceptible to dental caries than females, and gonadectomy reduced this susceptibility. To repeat these observations and to test the effect of testosterone propionate 55 young hamsters were grouped as follows: 12 control males, 10 castrated males, 5 castrated males implanted with 25 mg. pellets of testosterone propionate, 12 control females, 6 ovariectomized females, and 10 spayed females implanted with 25 mg. pellets of T. P. All animals were maintained on a high caries-producing diet for 121 days. When the teeth were examined for cavities, the caries incidence was so great in all animals that it was impossible to distinguish between groups. Average scores were: control males, 99; castrated males, 84; castrated males with T.P., 115; control females, 109; ovariectomized females, 107; spayed females with T.P., 76. These findings indicate that the sex difference in caries susceptibility in hamsters is best demonstrated when caries activity is not too great and that very careful control of all experimental conditions is necessary.

VII. Sixth Session: Sunday Morning, June 26; Abstracts 56-67

56. The Pattern of Resorption and Repair of the Root Surface. *Joseph L. Henry, and J. P. Weinmann, School of Dentistry, University of Illinois, Chicago, Ill.* This study is based on the microscopic examination of 15 human jaws containing 261 teeth and their supporting tissues. A resorption area was determined by the presence of a resorption or reversal line. The 922 resorption areas observed in these teeth were classified into active resorption, active repair, aplastic state, anatomic repair, functional repair, and alternating resorption and repair. The resorption areas were measured in length and depth as indices of their total size. Tabulation of the number, location, size, type, and age incidence of the resorption areas led to the following findings: (1) Over 90 per cent of the teeth showed histologic evidence of resorption of the root. (2) The number of resorption areas and the susceptibility to resorption increased with age. (3) Resorption areas were found more numerous on the mesial than on the distal surface, and also more numerous on the buccal than on the lingual surface. Little difference was noted between the right and left

or upper and lower teeth. (4) Repair was evident in 85 per cent of the resorption areas. Most of these were anatomically complete. (5) The average size of the resorption areas measured 0.73 mm. in length and 0.10 mm. in depth. This was found to be large enough to be visible in the roentgenogram under ideal conditions. (6) Seventy-seven per cent of the resorption areas occurred in the apical third of the root; 19 per cent in the middle third; and 4 per cent in the gingival third. The high incidence of resorption areas in the apical third may be explained by the fact that this area is largely covered by cellular cementum which is biologically more closely related to bone than is acellular cementum.

57. Genetic Hypo-Osteoclasia and its Effects on the Development of the Rat Incisor and Molar. *S. N. Bhaskar, I. Schour, R. O. Greep, and J. P. Weinmann, School of Dentistry, University of Illinois, Chicago, Ill., and Harvard School of Dental Medicine, Boston, Mass.* This study is based on the gross, roentgenologic, and histologic investigation of the dentition and the tibia of 72 animals of the *ia* strain of albino rats (Schour, Massler, and Greep, *J. Dent. Res.* **23**: 194, 1944), and 41 control rats varying in age from 18 days in utero to 307 days. The *ia* rats are characterized by a deficiency in the normal rate of physiologic resorption of bone and a consequent accumulation of bone tissue in the skeleton. The osteosclerosis in the tibia coincided with the most active period of its growth. It gradually regressed until 250 days of age when the bones appeared normal. Hypo-osteoclasia in the premaxilla, maxilla, and the mandible had far-reaching effects. The bone of the dental crypts failed to resorb in response to the enlarging tooth germs and the latter became distorted. The epithelium of the enamel organs proliferated into the bone marrow spaces and there led to the ectopic formation of enamel, dentin, and cementum. The deposition of hard dental tissues between and on the bone trabeculae led to ankylosis, failure of eruption of teeth, and the formation of odontomes in the region of incisors and molars. Because of the different growth potentials of the incisor and molar tooth germs, the odontomes associated with the incisors continued to increase in size with age while those of the molars were limited in their growth. Some of these findings resemble those reported by Grüneberg in the grey lethal mouse (*J. Anat.* **71**: 236, 1937).

58. Three Dimensional Animal Cephalometry. *Joseph R. Jarabak, D.D.S., M.S., and John R. Thompson, D.D.S., M.S.D., M.S., Northwestern University Dental School, Chicago, Ill.* Three dimensional animal cephalometry was the outgrowth of a need for a means of study of facial asymmetries in long snouted animals, such as rats and dogs. The method of orientation of the animal for anteroposterior and lateral cephalometric roentgenograms has been described previously by Jarabak. For the occlusal or third dimensional method of orientation, the animal is placed into the cephalometer with the ear posts entering into the uppermost portion of the external auditory meati. The rostral portion of the skull is extended in a line parallel to the long axis of the animal, viz., in the vertical plane. The point of entry of the central ray is at right angles to Frankfort horizontal plane or through the superior surface of the cranium at the level of the external auditory meati. The film is placed in the front of the inferior surface of the mandible at a constant distance caudocephatically and anteroposteriorly. Regular $2\frac{1}{4} \times 3$ inch roentgen film without intensifying screens was found to give the best results.

59. Effect of Unilateral Resection of Facial Nerve on Facial Growth of the Rat. *Joseph R. Jarabak, D.D.S., M.S., Maclyn Kamins, M.D., and Karl Vehe, M.D., Northwestern University Dental School, Chicago, Ill.* The facial nerve was resected at the stylomastoid foramen. Resection at this level causes

a paralysis of all of the muscles of the face. Upon recovery from the anesthetic there is a very slight rostral deviation to the side opposite of the resection. The rats are cephalometrically appraised monthly for 12 months. It was observed that the rostral deviation becomes more pronounced monthly for 4 months and thereafter remains unchanged. The lateral cephalometric plates indicate little or no change as compared with the controls. The change is readily seen in the anteroposterior and occlusal cephalometric roentgenograms which show a septal and rostral deflection to the opposite side of the resection. The bony structure of the face of the normal side appears smaller than that of the resected side. In the occlusal roentgenogram the rostral portion appears to deflect from the region of the malar process to the upper incisor region. The upper incisor in the anteroposterior roentgenograms appears to be on a higher plane than the incisor of the opposite side. There is also a change in the incline plane of the molars on the two sides.

60. Condylar Resection and its Effect on the Growth of the Face of the Rat. *Joseph R. Jarabak, D.D.S., M.S., Karl Vehe, M.D., and Maclyn Kamins, M.D., Northwestern Univerity Dental School, Chicago, Ill.* Immediately following the resection and removal of the rat condyle there is a deflection of the mandible to the side of the condylectomy. Within 10 days the progressively growing incisors lock the mandible in malocclusion with the maxilla when the jaws are closed. In 5 weeks the upper incisor on the side opposite the resection forms a complete circle and the incisor on the resected side begins to enter into the palatine foramen. The lower incisor on the resected side continues to grow and eventually pierces the upper alveolar ridge. The lower incisor of the opposite side contacts the upper incisor and wears accentrically into a needlelike structure. The molars which have been taken out of normal function adjust themselves into occlusion by elongation and tipping. The attrition on these teeth indicates that there has been a complete change in the adaption of molars to the existing malocclusion. In the alizarinated material there are indications of rapid growth activity in the resected stump as well as an increased growth in the lateral surface of the mandible opposite the mandibular foramen. There appears to be an increased activity in bone growth in the inferior border of the mandible distal to the alveolar process of the lower incisor. There is also an increase amount growth activity or growth readjustment in the temporal portion of the zygomatic arch.

61. Cephalometric Appraisal of the Cranium and Mandible of the Rat Following Condylar Resection. *Joseph R. Jarabak, D.D.S., M.S., and John R. Thompson, D.D.S., M.S.D., M.S., Northwestern University Dental School, Chicago, Ill.* Following the resection of the condyle there is a deflection in the mandible to the side of the resection, this is shown in both the anteroposterior and occlusal roentgenograms. The degree of deflection depends entirely on the amount of the neck of the condyle that is removed with the condyle. This immediate twisting is the response to a mechanical shortening of the mandible in the most potent growth area. In the occlusal and anteroposterior roentgenograms taken from the first month following resection through the fifth month, there was appreciable growth in the normal half of the mandible. The mandibular deflection does not appear any greater than immediately following the resection. In the occlusal roentgenogram the molars show considerable displacement or adjustment to the new mandibular position. In the anteroposterior roentgenogram it appears as though the whole alveolar process and the teeth have shifted to the side of the mandibular deflection. There also appears to be a twisting of the entire cranial base to the side of the resection. This was most

noticeable in the rats that survived beyond the fifth month. In the lateral roentgenogram there appears to be some discrepancy in growth between the two halves of the mandible. This discrepancy is not as great as was expected in view of the removal of the condylar growth center. The anthropometric findings seem to substantiate the roentgenogram interpretation.

62. The Effect of Pyridoxine Deficiency on the Jaws of Mice. *B. Levy, Washington University School of Dentistry, St. Louis, Mo.* Twenty-eight mice of strain C_{57} black were fed diets deficient in pyridoxine. One group received a deficient diet which contained 18 per cent casein; a second group received a deficient diet, the casein content of which was 54 per cent. Histologic studies of serial sections of the mandibular condyle and periodontal structures of both groups of animals were made after one, two, four, and eight weeks. In the condyle, growth of cartilage and formation of bone were inhibited and finally ceased. The effects of the deficiency were accentuated in the high protein pyridoxine-deficient group. The gingivae of the pyridoxine-deficient group receiving 18 per cent casein showed a progressive atrophy of the interdental epithelium with a superficial necrosis and mild inflammatory reaction in the animals deficient for eight weeks. The height of the interdental alveolar bone slightly diminished with increasing deficiency. In the pyridoxine-deficient groups receiving 54 per cent casein, the above changes appear earlier and are more severe. Superficial necrosis of the epithelium covering the interdental papillae occurs after only two weeks; while in the animals deficient for eight weeks, the entire interdental epithelium is necrotic and there is a dense round cell infiltration in the underlying connective tissue. The alveolar bone shows greater resorptive changes with evidence of cessation of new bone formation.

63. The Effect of X-Radiation on the Jaws of Mice. *M. S. Burstone and B. M. Levy, Washington University School of Dentistry, St. Louis, Mo.* Young mice of strains C_{57} black and Swiss were exposed to 5,000 and 1,500 roentgen units of x-radiation in the area between the external auditory meatus and the lower border of the mandible using 50 kv., 2 Ma., O filter, t.s.d. 20 mm., H.L.V. 0.3 mm. Al for 18 to 62 seconds. Animals were sacrificed at various intervals postirradiation and serial sections of jaws studied histologically. A group of animals 2 to 30 days old were exposed to 5,000 roentgen units. Those animals which were irradiated over a small area at the apex of the incisor showed developmental abnormalities such as spiked, malformed, or hypoplastic incisors or failure of incisors to erupt. Histologic examination of the jaws shows marked disturbances in dentinogenesis, amelogenesis, and osteogenesis. The odontoblasts are distorted, lose their normal columnar appearance, and appear as large polygonal cells. There is cessation of normal dentin apposition with osteodentin formation throughout the pulp chamber. Many of the abnormal odontoblasts are trapped in this osteodentin or dentinoid material. The ameloblasts are destroyed in some areas; in other areas they appear as squamous epithelium, while in still other areas they are not affected. In areas where ameloblasts are disturbed, the enamel matrix is poorly formed or absent. There is fibrosis and aplasia of the marrow. Administration of 1,500 r to a second group of 3-day-old mice caused a hemiatrophy of the mandible, noticeable after 2 weeks. This results in occlusal imbalance. There is retardation of rate of eruption of the incisor on the irradiated side. Microscopic changes were similar to above, but were much less severe. In all animals, the irradiated side showed the more marked changes.

64. Calcium Exchange in Fresh and Unashed Bone and Dentin. *Elizabeth E. Underwood and Harold C. Hodge, University of Rochester School of Medicine and Dentistry, Rochester, N. Y.* The mechanism by which radioactive calcium is transferred from solution to glycol-ashed bone has been established as one of exchange. The dental tissues, dentin and enamel, also show the typical exchange pattern, a rapid phase of uptake and then a plateau. Further work on exchange of phosphate has been done by Neuman and his group on unashed tissues. He has demonstrated that the glycol-ashing procedure seriously modifies the bone as to the magnitude and the rate of the phosphate exchange. Since a study of the unaltered tissue has shown the mechanism of phosphate exchange to be different from that of a shed tissue, we decided that a similar study should be done with radioactive calcium. Fresh bone was compared with glycol-ashed bone as to the uptake of calcium by the tissue, as well as the percentage of the tissue available for exchange. The fresh tissue shows a much slower reaction which proceeds for the entire time period studied—30 days—in contrast to the glycol-ashed tissue which shows a two-phase reaction, an initial rapid uptake, and a steady state that is maintained for the experimental time. Fresh dentin and glycol-ashed dentin show a similar picture in all respects. The fresh dentin takes up the Ca_{45} at the same slow rate for the whole 30 days, whereas the glycol-ashed dentin reached a plateau within the first few days. The percentage exchange of bone in comparison with dentin shows the same general relationship in the fresh state as in the glycol-ashed state. The fresh bone exchanges more than the fresh dentin and that in turn exchanges more than the fresh enamel.

65. A Tracer Study of the Mechanism of Caries Prevention by Iodoacetic Acid. *Paul R. Thomassen and Henry M. Leicester, College of Physicians and Surgeons, San Francisco, Calif.* As part of a general investigation of phosphatase inhibitors and caries prevention, radioactive iodine was synthesized into iodoacetic acid which was fed or administered parenterally to young rats. After various periods of time, these rats were sacrificed and their teeth and other organs and tissues measured for radioactivity with the Geiger-Mueller counter. All the tissues examined, including the teeth, showed evidence of uptake of radioactive iodoacetic acid to various degrees. Radioautographs of rat incisors and molars showed deposition of active material on enamel, dentin, pulp, and supporting bone. The concentration was heaviest at the enamel surface and next to the pulp. This activity was of a transitory nature, and suggested that the uptake was an adsorption phenomenon rather than an incorporation into the apatite lattice as occurs with fluorine.

66. The Penetration of Labeled Phosphate From Silicate Into Dentin. *R. L. Glass and H. A. Zander, Tufts College Dental School, Boston, Mass.* Although several investigators have demonstrated the irritating properties of silicate cements, the cause of the irritation has not been definitely established. Most hypotheses center about the acidity of the hardened silicate. To determine whether or not phosphate from the silicate penetrates the dentin, cavities were prepared in dog teeth and filled with silicate cement containing P_{32} in the H_3PO_4 as a tracer element. The dogs were sacrificed from two to ninety-six hours after placing the silicate. The radioactive silicate was then entirely removed from each cavity preparation. The presence or absence of radioactivity in the dentin was determined by making radioautographs from thin sections of the teeth, and by testing the dentin and pulp below the cavity preparation with a Geiger-Müller counter.

67. Use of Radioactive Iodine as a Tracer in the Study of the Physiology of Teeth, II. Uptake by Human Teeth. *Herbert J. Bartelstone, Montefiore Hospital, New York, N. Y.* Observations were made on a series of 10 teeth extracted from patients receiving therapeutic doses of I^{131} using the technic described in *Science* (**106**: Aug. 8, 1947). Radioautographic technics utilizing no-screen x-ray film and Kodak medium lantern slide plates were employed to determine the distribution of the radioactive iodine 12 to 24 hours after the systemic introduction of the radioactive iodine to the patients. Where indicated, Grenz-ray examination of the sections were made and correlated with the morphological and radioautographic findings. There was uptake of I^{131} in the enamel, dentin, and cementum, with variations in degree and distribution. In the enamel there was marked uptake (a) at the surface, (b) in the subsurface enamel, (c) on the surface and in the subsurface of abraded enamel, (d) in calcium poor areas, and (e) in areas of marked organic material both surface and subsurface. In the dentin there was a significant increase over basal uptake in areas of metamorphosed and secondary dentin as well as carious, decalcified, and interglobular dentin. Many of the findings presented are not in accord with data previously published on the physiology of the hard structures of teeth.

IX. Seventh Session: Sunday Afternoon, June 26; Abstracts 68-77

68. A Preliminary Report on the Effect of Maternal Riboflavin Deficiency of Rats Upon Bones of Their Offspring With Special Reference to Cleft Palate. *Wm. H. Bauer, Saint Louis University, Schools of Medicine and Dentistry, Saint Louis, Mo.* Borderline deficient female rats were obtained by a carefully used and constantly checked modified Warkany's diet deficient in riboflavin alone. These rats, after remaining in anestrus two weeks, were given orally the minimum amount of riboflavin required to again start the estrus cycle. Such borderline riboflavin deficient rats were mated with normal male rats. Twenty-five per cent of thirty litters (25 per cent of 155 offspring) displayed various skeletal anomalies. Among these were 34 (22 per cent) cleft palates without harelips. These were always associated with an extremely reduced mandible and protruding tongue but normal-sized maxilla. The bones and endocrines of the rats and their offspring were studied microscopically. Since the cleft mainly involved the posterior part of the palate, seldom extended to the incisive papilla, and never involved the alveolar processes of the anterior parts, it evidently developed upon fusion of these parts. Under normal conditions the palatine shelves fuse when the tongue is withdrawn concomitantly with the drop of the growing mandible. The following working hypothesis is offered: In the offspring of borderline riboflavin deficient rats there is an interference with the endochondral ossification which affects the condyle thereby leading to a very marked shortening of the mandible. This dwarfed mandible fails to drop, the mouth remains closed, and the normally developed tongue interrupts the fusion of the palatine processes.

69. Lupus Vulgaris of the Oral Cavity. *Virgil D. Cheyne, D.D.S., Ph.D., School of Dentistry, University of Washington, Seattle, Wash.* Oral tuberculosis is usually a secondary involvement from advanced pulmonary lesions. Because it is accompanied by loss of resistance and advanced physical disability the commonly observed course is that of extensive ulceration with little or no healing or change in the progressive picture. In contrast, lupus vulgaris of the oral tissues, which is not accompanied by generalized debilitation, offers the opportunity to study chronic oral tuberculosis. A case is reported of a 9-year-old girl

who was first seen with tubercular lesions on her chin, left axilla, foot, and fingers which had existed for eight months. These were vesicular, scaly, and crusting, with little tendency to form large ulcerations. Biopsy sections showed the typical picture of tuberculosis. The lungs and oral cavity were grossly negative. Four months later a bright red lesion appeared on the upper right gingivae. It was not painful or ulcerated. Six months later two more lesions were present in the oral cavity, one near the upper left cuspid and another near the lower right bicuspids. During the next two years the patient was watched closely for new lesions and spread of old ones. Scarring appeared in healed areas of the mouth and it was noted in radiographs that progressive bony recession invited pocket formation with pus. Calculary deposits were consistently absent. At one time a large circular ulcer appeared in the palate, but this healed without scarring in two months. Various forms of local therapy were tried all during the observation period and the patient was hospitalized for long periods without success. The skin lesions continued to spread in certain areas, heal in others, and new ones appeared. At the time of the last observation four years from onset the skin lesions in general were unchanged. The oral cavity showed severe effects due to the disease.

70. The Effect of Estrogenic Stimulation on the Alkaline Phosphatase Activity of Oral Epithelium in the Rat. *J. R. Ring and B. M. Levy, School of Dentistry, Washington University, St. Louis, Mo.* Morphological changes (primarily hyperplasia and hyperkeratinization), associated with estrogen administration, have been described in the oral epithelium of women and female monkeys. In this investigation, no constant differences in structure were seen in routine preparations of oral mucosa from female rats (Wistar) sacrificed in diestrus, proestrus, or estrus, nor from spayed rats or spayed rats administered a massive dose of estradiol benzoate. Alkaline phosphatase preparations of oral mucosa, however, demonstrated an activity of this enzyme increasing from essentially negative during diestrus (and in the ovariectomized animals) to moderately strong in the oral epithelium of the estrous (and estrogen-treated) females. Phosphatase activity was negligible in the gingival epithelium (incisal papilla) even when it was marked in the contiguous oral mucous membrane.

71. Interpretations of Morphological Units of the Human Dentition. *A. A. Dahlberg, Department of Anthropology, University of Chicago, Chicago, Ill.* The key to interpretation of morphological units of the human dentition is to be found in the fact that there is greater stability of form, size, pattern, and expression in the first incisor, canine, first premolar, and first molar than in the posterior units of these respective tooth groups or series. Nutritional and certain pathological and environmental factors may readily be ruled out in these interpretations because they are rarely selective as to site in the dentition. Genetic and other anomalous characters are selective. Anomalous cusps and evolutionarily recent characters are recognized by their occurrence on the more variable teeth, principally the second incisor and second and third molars. Evolutionarily primitive characters are to be found more commonly on the morphologically stable teeth, that is, first incisor, canine, first premolar, and first molar. Therefore, it seems probable that the posterior members of each tooth group or series presents the possible future direction of the evolution of each of the respective tooth series. A knowledge of paleontology and comparative anatomy are essential in interpretations, particularly in distinguishing between homologous and analogous characters. Theoretically, mutations of genes which have a role in the determination of tooth form may occur in any direction, but most of the mutations which have occurred and persisted appear to fall within a rather limited range. The only real excep-

tions are those which have produced hereditary opalescent dentin and those which have resulted in complete anodontia. Other characters and conditions in the dentition must be studied further before they can be labeled the result of mutation of specific "genes of the dentition." Many characters spoken of in such manner are the result of suppressing or inhibiting factors resulting from correlative actions from other genes. Most instances of congenital absence of teeth and peg-shaped lateral incisors are in this category. In the long view of evolution only the small mutants producing slight variations are of any consequence. Interpretations of characters in the light of the extreme stability and expression of the key teeth and of the variability of their posterior members gives us the basis for attempts to solve many unanswered questions about the dentition.

72. Studies on Dental Paleopathology. II. Differential Diagnosis of the Post-Mortem Histopathology of Ancient Teeth. *Reidar F. Sognnaes, L.D.S., Ph.D., Harvard School of Dental Medicine, Boston, Mass.* Ancient teeth from various parts of the world have been sectioned by a plastic petrification method, described eleswhere (Sognnaes, Anat. Rec., Nov., 1947). Before exploring their developmental histological structure and intra vitam pathology, it proved necessary to identify various post-mortem changes on a microscopical level. The two most frequent post-mortem destructions take the form of either large lucunar surface decalcifications, or invasion and channeling of the teeth by certain types of fungi. The latter destructive agents do not destroy the enamel but invade the dentin and cementum without any "respect" for morphological pattern. Occasionally, all of the collagen-containing tooth substance may thus be destroyed, leaving only the shell of enamel intact. Differential diagnosis of these post-mortem changes requires microscopic examination which will reveal discontinuity that is produced in the developmental growth lines and normal morphology of the tooth, and absence of intra vitam reactions to the post-mortem destruction.

73. The Origin of Expansion in Orthodontic Treatment. *George B. Denton, Northwestern University Dental School, Chicago, Ill.* Contrary to statements made by Angle, Weinberger, and others, Fauchard did not introduce the expansion arch and he did not employ the principle of expansion. He did use a short metal strip to move teeth, and this device was extended by Bourdet. None of the eighteenth century orthodontists used an expansion arch, nor attempted to expand the arch. In the eighteenth century, instead of accommodating the arch to the teeth, practitioners always accommodated the teeth to the existing arch. The notion of widening the arch in the canine region was conceived by John Hunter, and he suggested a device—not an expansion arch—that would accomplish the result. Delabarre foreshadowed the objective of expansion early in the nineteenth century. Schange conceived it more clearly; Lefoulon early in the forties designed an appliance to expand the arch.

74. A Method for Measuring Vertical Masticatory Forces. *A. H. Howell, Engineering Department, Tufts College, Medford, Mass., and Finn Brudevold, Tufts College Dental School, Boston, Mass.* A method for measuring the vertical forces used during actual chewing of food has been developed. The principle involves a variable inductance type of strain gauge which is built under an artificial tooth to form a bite element. The bite element can be inserted in a denture or a removable bridge and is connected to associated electronic equipment and a strip chart recorder by means of thin wires. The forces are recorded as deflections on the strip chart, and measurements can be taken without interfering with normal chewing or the comfort of the test

subject. The masticatory forces were measured in a 55-year-old male who was wearing dentures. The mean and maximum force used during chewing of a mouthful of peanuts, cocoanut, and seeded raisins were recorded for the first and second premolar and the first molar. The masticatory forces were similar with different foods. The distribution of the forces on the teeth suggest that the brittle food, peanuts, was chewed mainly on the second premolar and the first molar, and that the tough foods, cocoanut and raisins, were shifted forward during mastication from the first molar toward the first premolar. The occlusal contact area was measured, and the first molar was found to have more than double the contact area of the two premolars put together. The pressure on the occlusal surfaces was calculated and appeared to be highest on the second premolar and the first molar when peanuts were chewed. When cocoanut and raisins were chewed, the pressure increased on all the teeth, but the maximal pressure was found on the first premolar.

75. The Effect of the Topical Application of Sodium Fluoride on the Enamel Surface. Microhardness of Vital Teeth. *A. B. Hord, D.D.S., and R. G. Ellis, D.D.S., M.Sc. (Dent.), University of Toronto, Toronto, Canada.* Once each week for 8 weeks the teeth of one side of each of three 5-month-old dogs were dried and painted with 2 per cent aqueous solution of sodium fluoride, which was allowed to remain on the teeth for 10 minutes. At the end of 8 weeks the animals were sacrificed and their extracted teeth immediately placed in 10 per cent formalin. Knoop hardness numbers for the treated and untreated teeth were determined using a Knoop indentor in a Tukon testor. In every instance the hardness of the enamel surface of the treated teeth was greater than that of the untreated teeth. The increase was at least 13 per cent in every case and in two instances 42 per cent.

76. Distribution of Radioactive Gallium in the Teeth and Jaws of Experimental Animals. *James A. English and Horace C. Dudley, Naval Medical Research Institute, Bethesda, Md.* Radiogallium in the form of soluble gallium lactate (pH 7.0 to 7.4) was injected subcutaneously into rats and rabbits. The dosage was 0.2 to 0.4 millicurie of Ga^{72} per kilogram of body weight (6 mg./kg.) which is well below the toxic level. The animals were killed in six hours and thin sections (50 to 100) were made of the bone and teeth by cutting with high speed abrasive discs and grinding to the desired thickness against an abrasive wheel. These thin sections were placed next to special roentgenograph film for 24 to 48 hours. The developed negatives were compared with transillumination photographs which were subsequently made from the ground sections. By this means it was determined that radiogallium is predominantly deposited in bone marrow, periosteum, tooth pulp, and periodontium. There is a moderate deposition in cancellous bone. Cortical bone and the dentin of the root portion of teeth take a barely perceptible amount of radiogallium, whereas coronal dentin and enamel give no indication that any radiogallium is absorbed. The main advantages of this method over those using the electroscope or Geiger counter for determining the radioactivity are that fractional portions of teeth and bone are not required, and the visual localization is rapid and effective. The relative distribution of the radioactive substance in each anatomic area is apparent; however, accurate quantitative measurements are not possible.

77. A Statistical Study of Bilateral Incidence and Extent of Dental Caries in White Rats. *Merrill G. Wheatcroft, Carl A. Schlack, Lincoln J. Gerende, B. L. Taylor, and V. J. Berzinskas, Dental Division, Naval Medical Research Institute, Bethesda, Md.* Diversified opinions may be obtained from a review of the literature on the subject of symmetry of bilateral incidence of dental caries in humans. It would be expected that bilateral symmetry of

incidence and extent of dental caries would exist between the grooves in the molar teeth of white rats. With this hypothesis in mind a statistical study was devised to determine the symmetry of incidence and occurrence of extent of decay. One hundred fifty-three males and 179 female, weanling, white rats from a controlled colony were fed an unlimited amount of a cariogenic coarse corn diet (HWC). The length of the experiment was 100 days after which the rats were sacrificed and their jaws prepared for study. The teeth were ground in an occlusocervical direction and under a magnification of ×10.5 the degree of decay of each groove scored. The data were compiled on master charts and a statistical analysis made. Bilateral symmetry of incidence of dental decay does occur between the grooves in the molar teeth of white rats in this experiment. The present study also shows, with a high degree of probability, that bilateral symmetry of the occurrence of extent of dental decay between grooves in the molar teeth of white rats does exist.

X. Materials Group Sessions: June 25 and 26; Abstracts 78-93

78. A Method for Determining Wear Resistance of Human Enamel and Artificial Teeth. *F. A. Slack, H. D. Justi & Son, Inc., Philadelphia, Pa.* A method has been devised which compares the wear resistance of human enamel, artificial teeth, or other like substances, to a standard. All of the objects are compared directly. Although the results seem to coincide with clinical observations, no attempt is made to correlate wear resistance with wear in vivo. For the determination of wear resistance, a standard instrument is used, though the procedure has been changed somewhat. The test used may be duplicated in any laboratory. Milligrams weight loss is the method by which abrasion loss is determined. All objects tested registered a weight loss of from zero to one hundred, thus simplifying the scale. The test repeats itself within narrow limits of weight loss, and each test requires less than five minutes.

79. Physical Properties of Gypsum Products Used in Dentistry. *D. A. Mahler, School of Dentistry, University of Michigan, Ann Arbor, Mich.* This study deals with the physical properties of plasters and dental stones, and the application of these results to actual dental usage. The physical property which has been deemed most feasible for study is the modulus of elasticity. This parameter correlates the stress to which the material is subjected to the corresponding strain or deformation which the material experiences. In the attempt to achieve accurate and understandable results, some very interesting properties of these materials were discovered and will be mentioned accordingly. Some work has been done on thermal expansion of these materials subjected to varying environmental conditions and these results and their possible applications will also be discussed in regard to their effects on the dimensional accuracy of dental appliances.

80. Properties of the New Silicate Cement: Diafil. *G. C. Paffenbarger, A. C. Swaney, I. C. Schoonover, George Dickson and G. F. Glasson, National Bureau of Standards, Washington, D. C.* The pertinent physical properties of Diafil as related to its use in dentistry were measured. Its stability under conditions of use, opacity, resistance to impact, hardness, compressive strength, resistance to staining, pH of aqueous extract, shrinkage during setting, and solubility were of the same order as other silicate cements on the list of Certified Dental Materials as established by the National Bureau of Standards and the Council on Dental Research of the American Dental Association. Many claims which the manufacturer of Diafil makes for it could not be verified in this investigation. The sample tested complied with the requirements of the American Dental Association Specification No. 9 for Dental Silicate Cement.

81. The Effect of Surface Contact in the Retention of a Denture. *E. W. Skinner and Paul Chung, Northwestern University Dental School, Chicago, Ill.* Baseplates in acrylic resin were constructed to fit a model, the surface of which was overlaid with an elastomer resin to simulate the soft tissues of an edentulous upper arch without undercuts. Distilled water was used as the intermediary liquid film. It was found that the presence of relief areas reduced the retention; the use of post dam and peripheral seal, either separately or together, increased the retention. The peripheral seal was as effective as the post dam in increasing the retention at the posterior border, but it was more effective in increasing the lateral retention. The ridge area at its height of contour contributed nothing to the retention of the denture baseplate.

82. "Self-Curing" Acrylic Resins. *P. B. Taylor, and Sybil Frank, Cosmos Dental Products, Inc., New York, N. Y.* The presentation is in the nature of a progress report upon "self-curing" acrylic resins. Included in the report is a discussion of the mechanism of polymerization, the problems involved in securing color fastness, discussion of molecular weight of polymer and degree of polymerization, and the comparison of some physical properties of normally and "self-cured" acrylics.

83. Some of the Properties of Acrylic Resins that Polymerize at a Comparatively Low Temperature in Relation to Their Use As a Denture Repair. *H. J. Caul, I. C. Schoonover, George Dickson, and G. C. Paffenbarger, National Bureau of Standards, Washington, D. C.* The strength, stiffness, and warpage of two brands of denture repair materials which polymerize at room temperature are compared with resins on the list of Certified Dental Materials.

84. How Strong is Dental Amalgam? *W. T. Sweeney, School of Dentistry, University of Michigan, Ann Arbor, Mich.* This report was a condensation of a rather extensive series of tests made on five typical dental amalgams to determine the effect of manipulation on the compressive strength. The methods of testing were discussed. The effect of size of test sample, rate of load application, methods of mixing and packing, etc., on the strength values obtained were covered in considerable detail. The results are based on tests of several hundred specimens prepared under controlled laboratory conditions. The rate of increase in strength in the early stages of setting, from twenty minutes to seven days after mixing, was included. The relation of stress to strain in amalgam was included, as well as the modulus of elasticity in compression. A method for the determination of the elastic and plastic range of amalgam alloys has been devised that should lead to new or slightly different methods of evaluating the strength of amalgam. Recommendations for more accurate defining of testing procedures were made so that better evaluations of the strengths of different alloys may be possible.

85. Strength of Amalgam as Related to Time. *R. W. Phillips, School of Dentistry, Indiana University, Indianapolis, Ind.* Six popular alloys were studied in order to determine their compressive strength as it is related to time. Crushing strength was tested at various time intervals ranging from one-half hour up to six months. It was found that amalgam, though quite weak during the first few hours, rapidly gains its strength and generally reaches approximately 85 per cent of its maximum by the end of the first eight hours. The compressive strength thereafter continues to increase gradually up to the sixth-month final test. Hardness measurements verified these findings. These crushing strength measurements can probably be associated with the setting reactions and indicate that equilibrium in amalgam is not reached for an indefinite period of time.

86. Physical Characteristics of Material in Relation to Structural Usefulness. *R. C. Brumfield, J. F. Jelenko & Co., Inc., New York, N. Y.* This paper notes the fact that the physical characteristics of many dental materials are today available to the dental profession which are both accurate and adequate. These physical characteristics have several indirect types of usefulness, but in order to be directly serviceable they must be applied to the intelligent structural design of those appliances which are used in the mouth. The three types of information necessary to such design of any structure are listed, and it is noted that only one of these three is at present available to dentistry. It is further shown that the availability of one type only is of very limited direct application. A plea is made for more basic education in dental schools in courses dealing with structural matters, and for more research as to the controlling forces, both in character, and intensity, which determine the structural design of various types of dental appliances. This general discussion is supplemented by a practical example of the application of structural laws to a specific dental structure (free ended lingual bar case under lateral occlusion). Methods are given for determining the moment of inertia, section modulus, and other characteristics of oddly shaped sections, and curved beam analysis is applied to the lingual bar. Conclusions are drawn which show a benefit to the dental profession of such analysis in guiding the intelligent design and construction of dental appliances.

87. Fiberglas in Dentistry. *Harry Maeth.* Fiberglas, which is glass in a fiber or filament form, has been observed in dental applications as a root canal filling material, as a temporary filling material, as a base under gold foil and amalgam restorations, and as a pulp chamber filling material in the treatment of deciduous teeth after pulpotomy. Fiberglas is an inorganic, nontoxic, nonallergenic, nonsensitizing, and chemically stable substance that produces no harmful effect upon human tissues. Glass is not classed as a carcinogenic substance. It is pliable and possesses great tensile strength and a high degree of dimensional stability. It is nonhygroscopic and noninflammable. The individual fibers do not absorb water; Fiberglas is therefore easily sterilized and resterilized. Glass fibers possess the greatest tensile strength-weight ratio of any commercial material, natural or synthesized by man. Fibers averaging 23/1,000,000 inch in diameter have a tensile strength of more than 250,000 pounds per square inch. The physical appearance of Fiberglas is not unlike that of white cotton wadding. The filaments are about 3 microns in diameter. In dental use Fiberglas serves as an insulating and a reinforcing material. The working mass, puttylike in consistency, which sets to a density closely comparable to that of dentin, contains zinc oxide, eugenol and/or beechwood creosote, a trace of rosin, a trace of zinc acetate, and the Fiberglas, hammer milled to $\frac{1}{16}$ inch length, superfine fiber, clean glass without treatment.

88. A Note on the Deterioration of Dental Impression Compounds. *A. R. Docking and Joan A. Donnison, Commonwealth Bureau of Dental Standards, Australia. (This paper was read by Claude Watts, Ransom & Randolph Company, Toledo, Ohio.)* Impression compound released for disposal from Air Force stores after holding for at least five years was found to have working properties inferior to fresh material. The compound in question was of the semielastic type and its ability to recover after deformation had obviously decreased. Laboratory tests confirmed that the flow characteristics had altered appreciably. Chemical examination of the old samples showed that, in comparison with fresh material, the acid value, acetyl value, and iodine value were lower, indicating some polymerization and particularly polycondensation. In

this instance the constituent most prone to alteration on storage was probably shellac. It was concluded that the "disposals" material was no longer suitable for use as an elastic type impression compound and that there was no economic method for recovery of the unaffected constituents.

89. Hygroscopic Expansion in Some Casting Investments. *N. O. Landgren, Dental School, Stockholm, Sweden, and School of Dentistry, University of Michigan, Ann Arbor, Mich.* This report covers an investigation of the hygroscopic expansion for investment materials available in the market. The investments include Ransom & Randolph Hygroscopic, Whip-Mix Beauty Cast, Ransom & Randolph Gray, and Kerr Cristobalite. The method of determining expansion was that described by Skinner and Degni in which the investment was allowed to set in an inlay casting ring. Dimensional change was measured by the use of a dial micrometer placed in contact with the investment during the setting. Results indicate that all four investments show hygroscopic expansion, and the amount of expansion varies with the time and temperature of immersion in water. Measurements were also made with variations in water/powder ratio, the use of one or more asbestos liners in the ring, and a change in temperature of the water in which the investment was immersed.

90. Low Heat vs. High Heat Processing for Chrome Base Alloys. *E. S. Griffiths, Ticonium, Albany, N. Y.* A step-by-step description of the procedure and technic employed in processing one chrome alloy (Ticonium) using a low heat technic and plaster-bound investment, and a discussion of the effect of mold temperature and casting temperature on the grain size of the casting and its physical properties was presented.

91. Some Physical Properties of Zinc Oxide-Eugenol Impression Pastes. *E. W. Skinner, H. W. Ziehn, and E. N. Cooper, School of Dentistry, Northwestern University, Chicago, Ill.* The setting times, hardness, and dimensional changes of a number of commercial brands of zinc oxide-eugenol impression pastes have been studied at the temperature of the mouth (37° C.) and at the temperature of the room (25° C.) The setting times of some of the pastes are radically affected by the temperature. Several of the pastes never hardened completely.

92. Vertical Dimension of Denture Base Materials. *Naomi S. Steck, Vernon-Benshoff Co., Pittsburgh, Pa.* A description of a method for measuring change in vertical dimension of denture base materials, and a discussion of the data obtained from this method, including change inherent in the denture materials as well as change due to the human factor involved in processing denture base resins, and a comparison of compression and injection molding as it affects vertical dimension were presented.

93. Some of the Factors that Influence the Polymerization of Acrylic Resins. *T. E. Fischer, I. C. Schoonover, George Dickson, and G. C. Paffenbarger, National Bureau of Standards, Washington, D. C.* The amount and kind of inhibitor, the presence of impurities and the conditions of storage of acrylic liquid (monomer) and acrylic powder (polymer) prior to use affect the properties of dentures made from these resins. Certain of the inhibitors employed produce a yellow color in the resin. Impurities prevent complete polymerization and produce color changes in the resin. Polymer powder stored at elevated temperatures may become unsuitable for use as a denture base.

XI. Papers Read by Title; Abstracts 94-131

94. Use of 9-Aminoacridine Hydrochloride Solution in Endodontia. *Lester E. Breese, University of California, College of Dentistry, San Francisco, Calif.* The drug was used for pulp canal and periapical sterilization in 31 cases, 20 completed to date. Methods used: (1) Placement of a dry paper point in the canal, saturation of the point followed by a pumping action; (2) fitting a hypodermic needle into the canal with a Luer-Lok syringe attached, sealing off with unvulcanized rubber, and applying the drug under pressure, and (3) anesthetizing the area over the periapical involvement and forcing a 20 gauge hypodermic needle through the cortical plate with a syringe attached. Aspirating, then replacing the first syringe with one containing the drug, and irrigating the area, exit via canal. Cases treated were diversified as to condition, most having extensive periapical involvements. The largest area was an acute periapical abscess of an upper lateral with a parulis forming on the palatal surface. Estimated size determined from the roentgenogram was 15 × 17 mm., extending 5 mm. along the root, displacing the cuspid. Drainage to reduce inflammation, followed by 4 penicillin in oil applications using method (2) given one day apart, resulted in a positive culture. One 9-aminoacridine hydrochloride treatment, using method (3) gave a negative culture. A 14-month roentgenogram checkup showed bone repair except for a 1 × 1 mm. area at the tooth apex. Ten other cases averaging 6.5 × 8.6 mm. areas with a 4.8 mm. root involvement show increasing bone repair with each recall for evaluation. Nine cases have not been checked since completion. The drug was used after others had failed, more often then it was used exclusively. One case of edema occurred following injection into the periapical area, however, no pain was experienced. An allergy to the drug or to procaine was suspected.

95. Dehydrogenases in Saliva. *J. C. Calandra and L. S. Fosdick, Northwestern University Dental School, Chicago, Ill.* It was recently shown that the reductase activity of saliva is a function of caries activity. The method of determining the dehydrogenase activity gave no indication as to the specific dehydrogenases present. This paper describes an attempt to determine the source of dehydrogenases in saliva. The method used was to test the oxygen uptake of specific substrates in the presence of saliva. To date the following substances have been shown to be active substrates: glucose, hexose diphosphate, lactic acid, dl-glyceric aldehyde, d-glycine, d-methionine, l-cystine, d- and l-norleucine, d-alanine, d-valine, l-histidine, l-arginine, l-glutamic acid, dl-lycine, dl-aspartic acid, sodium pyruvate, sodium stearate, and sodium palmitate.

96. The Effect of Water-Soluble Chlorophyll "A" on Wound Healing. *M. E. Chapin, D.D.S., and G. W. Rapp, Ph.D., Loyola University School of Dentistry, Chicago, Ill.*

97. Further Studies on the Effect of Human Saliva on the Cholera Vibrio, in Vitro. *C. E. Dawson and W. Blagg, Dental Division, U. S. Naval Medical Research Unit No. 3, Cairo, Egypt.* Previous studies revealed evidence that saliva from normal healthy individuals contained one or more agents which exhibited antibacterial properties on the cholera vibrio in vitro. Saliva from a cholera-infected group showed little or no antibacterial properties against the test organisms. In general, cholera infection with high morbidity was limited to the malnourished classes. These correlations prompted analysis into the antibacterial effectiveness of saliva from healthy and malnourished individuals. Stimulated and unstimulated saliva was collected from 20 healthy and 10 mal-

nourished subjects, and filtered by means of aluminum oxide, glass wool adsorption columns. By a method which makes use of spectrophotometry and plating for determining the antibacterial powers of saliva, results were obtained which indicated that saliva from normal healthy individuals contained at least two active principles capable of inhibiting and lysing the cholera vibrio. These active principles could not be demonstrated with the same degree of potency in malnourished individuals or acute cholera patients. It is felt that the antibacterial titer of saliva might serve as an indication of the effectiveness of other body fluids, namely intestinal secretions. The findings offer a possible explanation as to why acute symptoms of cholera are for the most part confined to the malnourished peoples, and the control of cholera is more of a social and economical problem than a medical one.

98. A Histological Study of the Relationship of the Root of the Mandibular First Molar of the Rat in Growth and Eruption to the Lower Border of the Mandible. *Benjamin Dienstein and James Nuckolls, University of California, College of Dentistry, San Francisco, Calif.* The mandibles of ten rats ranging in age from birth to 31 days were serially sectioned as uniformly as possible in buccolingual aspect through the mesial root of the first molar. Measurements were taken from photomicrographs of these sections and growth curves and growth-ratio curves were plotted to determine how the apex of the root was maintained spatially in the growing mandible during the growth and eruption period. The apex of the root was found to move away from the lower border when the lower border was considered a fixed point. When the movement of the apex was related to other points on the growing tooth and mandible, it was found to maintain a discrete position with regards to the lower border. Eruption of the tooth starts when the root of the tooth begins to form.

99. Pathologic Pigmentation of the Oral Mucosa. *Clifton O. Dummett, D.D.S., M.S.D., M.P.H., School of Dentistry, Meharry Medical College, Nashville, Tenn.* Pigmentation of the oral tissues is observed in a large number of individuals with no oral pathosis. It may also occur as a result of disease, so that in oral diagnosis it is necessary to differentiate between normal and abnormal pigmentations. Observations were made on patients presenting at the dental and medical clinics of Meharry Medical College over the past six years and cases were selected in which there were pigmentary changes resulting from various abnormal conditions. These pigmentations fell conveniently into two categories on the basis of their local or systemic etiology. Of local origin were the pigmentations resulting from the inflammatory effects of irritants which may be mechanical, atmospheric, chemical, infectious, or traumatic. In addition, irritation may also result from roentgen-ray irradiation, food impactions, food stains, or from calcareous and other dental deposits. Of systemic origin were the pigmentary changes, much more complex in etiology, resulting from systemic conditions which produce oral manifestations. Some endocrine diseases, the metallic stomatites, dietary deficiencies, the blood dyscrasias, pregnancy, and drug allergies are among the commoner conditions indictable in oral pigmentogenesis.

100. Oral Capacity Index. *Clifton O. Dummett, D.D.S. M.S.D., M.P.H., and Theodore E. Bolden, D.D.S., School of Dentistry, Meharry Medical College, Nashville, Tenn.* It has been thought necessary to include among the established criteria of oral normalcy the added criterion of "oral capacity." In this study, measurements were made of the total oral fluid capacity which may be defined as the maximum volume of water which can be introduced into the mouth at a

particular time. This estimate has been designated as the O.C.I. (oral capacity index) and would vary according to age, sex, race, typal form of face, oral habits, strength of oral musculature, number of teeth, and presence of dental prosthesis. These studies purport to measure the O.C.I. under these conditions and then to determine what effects the above-mentioned factors would have on this index. A further anthropological extension of the study is in progress to determine whether any relationships exist between oral capacity and external orificial measurements. By means of the orocapacitometer, O.C.I scores in seventy-one subjects were obtained. These scores varied from a low of 76 c.c. for an 8-year-old girl with square facial form, to a high of 228 c.c. for two male subjects, one 28 years of age with tapering facial form and the other 22 years of age with square facial form. O.C.I. scores of three subjects who were wearing prosthetic replacements decreased from 5 to 17 c.c. when the appliances were in place.

101. Quantitative Comparisons of the Roentgenographic Resistance of Negro and White Patients Insofar as These Resistances Concern Dental Roentgenography. *Clifton O. Dummett, D.D.S., M.S.D., M.P.H., School of Dentistry, Meharry Medical College, Nashville, Tenn., and Albert G. Richards, B.S., M.S., School of Dentistry, University of Michigan, Ann Arbor, Mich.* Some dental roentgenologists believe that dental roentgenograms of Negro patients should have a longer exposure time than roentgenograms of white patients of similar size, age and sex, in order to produce films of a desirable density. Although it is believed that the need for additional exposure is because of the fact that the oral tissues of the Negro are more resistant to the passage of roentgen rays than those of a corresponding white person, no adequate studies have been done on this particular phase of the subject. If additional roentgen-ray resistance in these oral tissues does exist, it could be due to the amount of pigmentation, the thickness and consistency of the soft tissues of the cheek, the size, composition and amount of calcification of the teeth, the thickness of the lingual and buccal or labial plates of bone of the maxilla and mandible, and the entire thickness and density of these bones. Investigations are in progress to compare quantitatively the roentgen-ray resistances of Negro and white patients insofar as these resistances concern dental roentgenography. Up to this time, findings on a small number of patients indicate that Negro patients have not appeared to be any different from the white patients insofar as soft tissue radiopacity is concerned. Furthermore no difference has been noted between Negroes of varied complexions.

102. Microscopic Carieslike Lesions Produced in Vitro in the Enamel of the Syrian Hamster. *Valerie Hurst, James Nuckolls, and Harry E. Frisbie, University of California College of Dentistry, San Francisco, Calif.* The ability of oral and extraoral bacteria to produce microscopic carieslike lesions in hamster enamel in vitro is being tested by the method previously described. Thirteen strains of oral actinomycetes, 14 strains of oral lactobacilli, 4 strains of alpha streptococci, 4 strains of oral diphtheroids, 3 strains of *Corynebacterium diphtheriae*, and 7 different bacteria of extraoral origin, are being tested on more than 500 unerupted hamster molars in the experiments now in progress. Similar tests on 23 unerupted human molars are also now in progress. Histologic examinations of approximately 100 of the hamster molars are now completed with the following results: Lesions have been produced by 10 out of 13 oral actinomycetes in 22 out of 29 molars incubated in the broth cultures for 65 to 161 days at neutral pH. No lesions developed in 10 molars incubated 15 to 46 days under these conditions. Lesions have been produced by 3 out of 6

oral lactobacilli in 4 out of 9 molars incubated in broth cultures for 65 to 130 days at neutral pH. A minute probable, but not definite, lesion developed in one out of 5 molars incubated for 15 to 45 days under similar conditions. The enamel was almost entirely dissolved from 5 molars incubated in dextrose broth cultures of the same lactobacillus strains for 51 to 130 days at pH 3.4 to 4.0. Their appearance was unlike the histopathologic picture of caries. Lesions were present in the undissolved enamel matrix of 4 molars incubated in the dextrose broth cultures for only 15 to 45 days at pH 3.9 to 4.2. No lesions were found in the undissolved matrix of 3 molars incubated in malt broth lactobacillus cultures for 32 to 64 days at pH 3.2 to 3.4 Non-nutrient washed cell suspensions of the actinomycetes and the lactobacilli have not produced lesions in the few tests completed thus far. No lesions have been produced, as yet, by any of the other bacteria tested.

103. Sugar Utilization by Oral Actinomycetes and Lactobacilli. *William E. Hutton, Valerie Hurst, and James Nuckolls, University of California College of Dentistry, San Francisco, Calif.* Pure cultures of actinomycetes and lactobacilli known to produce carieslike lesions in molars of the Syrian hamster in vitro were tested for the utilization of certain sugars. The medium was a peptone (2 per cent)- dipotassium phosphate (0.5 per cent)- sugar (2 per cent: omitted in controls) broth (incubation: candle jars at 37° C.). Titratable acidity, terminal pH (Coleman pH electrometer) and, in certain experiments, the amount of cell material produced (dry basis) as mg. per ml. of medium were determined. Each of seven actinomycete strains and each of three lactobacillus strains fermented glucose, maltose, and sucrose, while only two strains each of the lactobacilli and actinomycetes fermented lactose. In 30 hours' incubation, (initial pH range 7.6 to 7.8) six of seven actinomycete strains produced heavier growth in glucose peptone broth than in peptone broth. Three of seven actinomycetes strains produced average growth of 0.3 mg. per ml. while three of three lactobacillus strains produced average growth of 0.1 mg. per ml. in glucose peptone broth. In seven days, seven of seven actinomycete strains produced from sucrose an average of 0.03 milli-equivalents of acid per ml. of medium as opposed to 0.006 meq. per ml. for three of three lactobacillus strains. Terminal pH ranges were: actinomycetes, 5.0 to 5.8; lactobacilli 6.5 to 7.6 (initial pH, 7.8). The data show that under certain conditions, certain actinomycetes were able to produce more acid from sucrose and heavier growth from glucose than were certain lactobacilli. It is suggested that actinomycetes may be important in acid production in plaques on the surfaces of teeth.

104. Gingival Disease in Postwar Poland. *Irving Glickman, Tufts College Dental School, Boston, Mass.* The gingival condition of 450 males and females (ages 3 to 62 years) was examined in different areas of Poland in the first peacetime summer (1946) following seven years of war. For purposes of description, gingival disease was divided into three groups as follows: (1) Marginal, changes limited to the gingival margin; (2) diffuse, involvement in equal degree of the entire gingival mucosa including the gingival margin; (3) combined, diffuse involvement with accentuated marginal changes. The following prevalence of gingival disease was noted: marginal, 39.1 per cent; diffuse, 23.6 per cent; combined, 16.4 per cent. Total gingival disease, 79.1 per cent. Gingival disease increased with age as follows: 3 to 6 years, 28.6 per cent; 7 to 14 years, 58.9 per cent; 15 to 20 years, 89.5 per cent; 24 to 40 years, 93.1 per cent; 41 to 62 years, 97.6 per cent. Only 4 cases of ulceromembranous gingivitis were seen in the entire group examined.

105. The O_2 Consumption of Gingival Tissue in Different Stages of Healing. *Irving Glickman, Samuel Turesky, and John Manhold, Tufts College Dental School, Boston, Mass.* A correlated histologic and manometric (Warburg technic) study was conducted of gingival tissue in various stages of healing following gingivectomy. The marginal gingiva in four areas in each of twenty-four dogs was removed and the different operated areas subsequently studied microscopically and manometrically at intervals up to sixty-four days after the initial operation. The oxygen consumption of normal dog gingiva was established at QO_2 1.4 with a standard deviation of 0.1. The findings in healing gingiva indicate the following: (a) healing was completed in 46 days after gingivectomy; (b) oxygen consumption increased in the early stages of healing associated with increased vascularity, formation of new connective tissue cells, proliferation of epithelium, and inflammatory changes. The oxygen consumption diminished in the direction of the normal value as healing approached completion.

106. Observations on Experimental Dental Caries. VII. The Influence of the Removal of the Cheek Pouches. *Humberto Granados, Johs. Glavind, and Henrik Dam, Polytechnic Institute, Department of Biology, Copenhagen, Denmark.* Thirty-two newly weaned male hamsters were litter-mate distributed into 2 groups (16 animals in each). The cheek pouches of the animals in Group 1 were totally removed by a technic previously described (Granados, *Odontologisk Tidskrift* **56**: 462, 1948). Group 2, control, was sham operated. Both groups were reared for 100 days on the following diet: yellow corn (finely ground) 25 per cent; sucrose 25 per cent; corn starch 20 per cent; powdered whole milk, 22 per cent; ether-extracted yeast, 5 per cent; alfalfa meal, 2 per cent; and salt mixture, 1 per cent. Each 100 Gm. of this ration were supplemented with 10 mg. of vitamin E. Both groups were given tap water ad libitum, and were weighed every week. These studies were carried out between the months of November and February. On completion of the 100-day experimental period the animals were killed and autopsied. The growth rate of Group 1 was slightly lower than that of Group 2 during the first seven weeks, thereafter being the same for both groups. The final weight was also the same in both groups. The caries activity, as measured by Keyes' method (*J. D. Res.* **23**: 439, 1944), was as follows. Group 1: average number of carious molars 6.8, average number of carious lesions 8.6, average caries score 4.9. Group 2: average number of carious molars 7.1, average number of carious lesions 8.7, average caries score 5.0. This experiment shows that the removal of the cheek pouches which the hamsters normally possess does not influence in any direction their dental caries activity.

107. Observations on Experimental Dental Caries. VIII. The Effect of Dietary Vitamin C. *Humberto Granados, Johs. Glavind, and Henrik Dam, Polytechnic Institute, Department of Biology, Copenhagen, Denmark.* The claims of a positive effect of vitamin C in controlling or arresting human dental caries (M. T. Hanke: Diet and Dental Health, Chicago, 1933, the University of Chicago Press) have not been substantiated (Grandison, Stott, and Cruickshank, *Brit. D. J.* **72**: 237, 1942). Considering these contradictory results we have carried out the following studies. Thirty-two newly weaned female hamsters were litter-mate distributed into 2 groups (16 animals in each), and were reared for 100 days on the following diets. Group 1 (control): yellow corn (finely ground), 25 per cent; sucrose, 25 per cent; corn starch, 20 per cent; powdered whole milk, 22 per cent; ether-extracted yeast, 5 per cent; alfalfa meal, 2 per cent; and salt mixture, 1 per cent. Group 2: acorbic acid, 0.5 per cent mixed

in 99.5 per cent of the diet fed to Group 1. Both groups were given tap water ad libitum. The studies were carried out between the months of November and February. At the end of the 100-day experimental period, the animals were sacrificed and autopsied. The caries activity, as measured by Keyes' method (*J. D. Res.* **23**: 439, 1944), was as follows. Group 1: average number of carious molars 7.6, average number of carious lesions 9.4, average caries score 5.3. Group 2: average number of carious molars 8.5, average number of carious lesions 10.4, average caries score 5.9. It is clear that the addition of vitamin C to the diet did not induce any decrease of dental caries activity. These results are in agreement with those of Grandison and his co-workers.

108. A Systemic Factor Contributing to the Mineral Tide in Human Saliva. *B. F. Gurney, M.S., Loyola University School of Dentistry, Chicago, Ill.* Evidence has been accumulated that the variations in calcium and phosphorus concentrations of human saliva are quite exaggerated under conditions of controlled and uncontrolled ingestion of salts of these metals. These same fluctuations in salivary concentrations can be experimentally produced by histamine injections under proper conditions. The relation of histamine to certain systemic factors such as gastric secretion is well known.

109. The Formation of Mantle Dentin. *J. T. Irving, Ph.D., M.D., School of Medicine, University of Cape Town, Cape Town, South Africa.* Experiments have already been reported (*Nature* **158**: 486, 1946) in which it was found possible to arrest organic enamel formation in the upper incisor of the rachitic rat by a period of dietary restriction. In some cases this hypoplasia was seen as an arrest of enamel formation followed by an immediate resumption of amelogenesis; in some, amorphous enamel matrix was deposited for a while; in others, amelogenesis stopped completely for an appreciable period. It was noted almost invariably that when enamel formation stopped completely, the calcification of the dentin abutting on the area of enamel lack, usually known as mantle dentin, was also arrested. In cases where even only small shreds of amorphous organic enamel matrix were present, calcification of mantle dentin took place. Various workers have shown that the organic enamel contains, as it matures, more and more of the elements of calcification (Weinmann, J. P., Wessinger, G. D., and Reed, G.: *J. D. Res.* **21**: 171, 1942; Deakins, M.: *J. D. Res.* **21**: 429, 1942). It appears possible from the above observations that the mantle dentin obtains its requirements of calcification elements, at least in rachitic animals, not from the odontoblasts, but from the enamel organ via the organic enamel. The dependence of amelogenesis upon the presence of functioning odontoblasts has been stressed by Huggins, C. B., McCarroll, H. R., and Dahlberg, A. A. (*J. Exper. Med.* **60**: 199, 1934) and by Wassermann, F. (*J. D. Res.* **23**: 463, 1944), but the problem here appears to be of a different nature.

110. A Bone Serial Sectioning Method. *Joseph R. Jarabak, D.D.S., M.S., Northwestern University Dental School, Chicago, Ill.* A method for serial sectioning of rat skulls orientated on planes identical with those used in animal cephalometry has been developed. The skulls are embedded in monimeric methyl methacrylate or copolimer allyl ester styrene compound. A box type frame has been designed around the walls of which are placed four blocks of methyl methacrylate (polymerized), ⅛″ in thickness; these are held securely in position by "butt" screws which come out from the metal frame in the center of each block of plastic. The two pieces of plastic which comprise the lateral walls of the container are 2¼″ × 1½″ × ⅛″; the anterior and posterior walls are 1½″ × 1½″ × ⅛″. Three plastic posts, ⅛″ round, enter into the container to support the skull on Frankfort horizontal plane. Two of the posts serve as ear posts and enter into the "ear holes" and the third enters into the container

through the front section at 90° from the two ear posts. The aluminum base which comprises the base of this container is prepared so that two guide pins center the block in the same position each time. The saws used for sectioning are set up in a gang fashion of twenty-five cutters. The cutters used are three and one-half inch high speed screw setting saws. The shaft on which the saws are mounted may be moved up or down. Lateral and cross movement is accomplished by the "cross feed" upon which the specimen mounted in plastic is fixed. Sections are from .010 of an inch to ⅛ of an inch. The polishing is done by an inverted aluminum polishing disk to which has been attached a felt cloth impregnated with Tripoli.

111. Occlusal Attrition of the Rat Molars Following Facial Nerve Resection. *Joseph R. Jarabak, D.D.S., M.S., Maclyn Kamins, M.D., and Karl Vehe, M.D., Northwestern University Dental School, Chicago, Ill.* Appraisal of cephalometric radiographs of rats following facial nerve resection indicates that there is a change in the plane of occlusion of the molar teeth. The occlusal plane on the resected side is higher with respect to certain established planes of orientation than the normal side. There is an increase in the buccal lingual inclination of the plane on the abnormal side. This increase in inclination is not due to an increase in the tipping of the molars but the increase in wear or attrition along a different plane than is normal for the rat. Conversally there appears to be a flattening of the plane buccal lingually on the normal side. The change in the occlusal plane of the molars of the two sides seems to indicate that there is some change in the mode of function in the grinding of the food of the animal. There was very little or no appreciable difference in the incisal attrition of the incisors, indicating the possibility of a compensatory action by the muscle transversus. This muscle is situated in the anterior portion between the two halves of the mandible, its principal function being that of holding the two halves of the mandible together in rest and in the function of prehension and grinding of food.

112. Anthropometric Appraisal of Condylectimized Mandibles. *Joseph R. Jarabak, D.D.S., M.S., and Elaine Pierson, Northwestern University Dental School, Chicago, Ill.* In this series of mandibles the measurements taken were on the condylectimized and the noncondylectimized halves of the same animal. The animals range from 1 week to 5 months following the condylectomy. Measurements between the following joints were taken, distal tip of angular process of the mandible to the crest of the lower alveolus. This measurement was not affected to any marked degree. The distance between coronoid tip and lower alveolus was slightly shorter on the side of the resection. The distance from coronoid tip to distal tip of angular process was also slightly shorter on the side of the resection. The distance from the superior border of the coronoid tip to the inferior border of the angle of the mandible increased in size over the normal side. This indicates an increase in the height of the ramus on the side of the resection. The distance from distal tip of the angular process to the mesial of the first molar has become shorter on the side of the resection. Measurements affecting the length of the mandible following condylar resection are shortened whereas measurements associated with height of ramus seem to increase in proportion on the side of the resection over the normal side.

113. Sporicidal Effects of Hot Silicone Fluid. *Holmes T. Knighton, Washington University, School of Dentistry, St. Louis, Mo.* Since silicone fluid has been suggested as a substitute for oil in the sterilization of instruments (Crowley and Ostrander: *Science* **108**: 542, 1948), comparative sporicidal

tests were made as previously reported for hot mineral oil (Knighton: *J. A. D. A.* **38**: 309, 1949). Results indicated that hot silicone fluid had the same relative sporicidal effect as reported for hot mineral oil. For example, both silicone fluid and mineral oil at 150° C. required from 30 to 60 minutes to destroy spores killed by water at 100° C. within 15 to 20 minutes.

114. A Method for the Study of Tissue Changes During Orthodontic Movement as Affected by Eliminating Occlusal and Other Muscular Forces. *Joseph P. Lazansky, M.D., D.M.D., and Ferdinand S. Hoffmeister, M.D., Tufts College Dental School, Boston, Mass.* A method has been devised for studying changes in supporting tissues of teeth undergoing orthodontic movement. The technic permits exclusion of forces of the lips, cheeks, tongue, and masticatory muscles. Tissue changes occurring with and without the above-mentioned forces can be compared. Extraction of opposing teeth was employed to eliminate the forces of the masticatory muscles during occlusion, and an acrylic box covering the appliance served to eliminate the forces of the muscles of the cheek, lips, and tongue. A push-coil spring was used to apply force between the molar and premolar teeth of dogs. In order to obtain proper controls, three animals were required. One animal was used to compare the effects of orthodontic forces when the occlusal forces were eliminated on one side of the dentition. A second animal was employed to compare the effects of orthodontic forces, in the absence of occlusal forces, when lip, cheek, and tongue force were also eliminated on one side. The third animal served to compare the influence of orthodontic force in the presence and absence of all other forces.

115. Congenital Mesodermal Dysplasia (Osteogenesis Imperfecta, Dentinogenesis Imperfecta, and Blue Sclera). *Maury Massler, College of Dentistry, University of Illinois, Chicago, Ill.* The relationship between osteogenesis imperfecta and dentinogenesis imperfecta (hereditary opalescent dentin) has been suggested by Roberts and Schour, 1939, Blattner, Heyes, and Robinson, 1944, and by Massler, 1945. The clinical relationship between osteogenesis imperfecta and blue sclera is well known. Sixteen cases showing osteogenesis imperfecta, dentinogenesis imperfecta (hereditary opalescent dentin), and blue sclera, either singly or in combination, were observed and the following findings obtained: (1) osteogenesis imperfecta, dentinogenesis imperfecta, and blue sclera are clinically and probably genetically related but each character may manifest itself singly as well as in combination. (2) When osteogenesis imperfecta occurred alone, it was mild in character and sometimes remained latent for long periods of time (Lobstein's syndrome). (3) When osteogenesis imperfecta occurred in combination with either blue sclera or dentinogenesis imperfecta the occurrence of fractures was much more frequent. Clinically, therefore, the absence of blue sclera and dentinogenesis imperfecta (especially the latter) in a given case of osteogenesis imperfecta offered a better prognosis than when they were present. (4) When all three characters appeared together, the manifestations were very severe and sometimes appeared before birth. The infant may be born with as many as 25 fractures of various long bones. Healing and refractures were common in these cases. (5) The clinical relationship between the three characters is clear. The term congenital mesodermal dysplasia is offered to designate their concurrence as a clinical syndrome. (6) In a given case of dentinogenesis imperfecta (hereditary opalescent dentin), the child should always be examined carefully for the occurrence of blue sclera and a tendency to fracture after relatively mild trauma. In two of our cases, actual fractures after mild trauma did not appear until some years after the original examination.

116. Prevalence of Rampant Caries in Teen-Age Children. *Maury Massler, College of Dentistry, University of Illinois, Chicago, Ill.* Rampant caries is seen principally in two age groups. The disease attacks the very young individual between the ages of 4 to 8 years, ravishing the primary dentition; and it is also encountered in the teen-ager (11 to 18 years) in whom it mutilates the newly acquired permanent dentition. The actual prevalence rate of rampant caries during these susceptible periods has not, to our knowledge, been investigated. The prevalence of rampant caries was investigated in 2,842 high school students (1,256 boys and 1,586 girls) ages 14 to 17 years. These children lived in an industrial community of upper middle income and enjoyed better than the average amount of dental care. (1) Rampant caries was found in 5.0 per cent of the 2,842 children ages 14 to 17 years. (2) The girls were more often affected by rampant caries than the boys. Of the total of 143 cases of rampant caries, 82 (5.17 per cent) were girls and 61 (4.85 per cent) were boys. (3) Rampant caries had a strong predilection for the ectomorphic (slender) type of individual; 105 of the 143 cases of rampant caries were thin, slender ectomorphic) children; 34 were muscular (mesomorphic), and only 4 were stocky or fat children (endomorphic).

117. The Normal Oral Flora as Shown on Blood Agar Plate Cultures. *Maury Massler and Herbert L. Blum, College of Dentistry, University of Illinois, Chicago, Ill.* Aerobic blood agar plate cultures were made of swabs taken from the anterior pillars of the fauces and lower anterior gingiva of 850 normal healthy adults (predominantly male) ages 18 to 30 years. These cultures revealed that the dominant organism present on the gingiva was the alpha streptococcus *(Streptococcus viridans)*. They grew in great profusion in 96 per cent of the cases examined and in many cases were the only organism present on the culture plate. Gamma streptococci (nonhemolytic streptococci), growing in relatively much smaller numbers, were found in 27 per cent of the cases while *Staphylococcus albus* (nonhemolytic staphylococcus) was found in relatively small numbers in 20 per cent of the cases. Pneumococci and micrococci were seen somewhat less often. Beta hemolytic streptococci were isolated from 9 per cent of the cases while *Staph. aureus* were obtained in 5 per cent of the cultures. These organisms were considered to be pathogens or potential pathogens. The normal aerobic gingival flora differed only slightly but significantly from the corresponding flora on the anterior pillars. The flora on the throat revealed a somewhat lesser number and frequency in the occurrence of greening streptococci (82 per cent) and an increased number and frequency in the occurrence of gamma streptococci (75 per cent) and *Staph. albus* (27 per cent). Beta hemolytic streptococcus and *Staph. aureus* occurred more frequently on the throat (14 per cent and 10 per cent respectively) than on the gingiva.

118. The Oral Flora in Diabetics. *Maury Massler, College of Dentistry, University of Illinois, Chicago, Ill.* The oral flora of 88 diabetics (controlled and uncontrolled) ranging in age from 6 to 40 years was investigated by culturing the swabbings from the anterior pillars and lower anterior gingiva on ordinary blood agar plates. Hemolytic colonies (*Staphylococcus aureus* and beta hemolytic streptococci) were obtained on 57 (65 per cent) of the cultures. *Staph. aureus* was obtained on culture from the throat and gingiva of 40 (45 per cent) of the patients. Beta hemolytic streptococci were revealed on cultures from 35 (40 per cent) of the patients. Since the frequency of occurrence of these organisms on the throats and gingiva of 850 normal healthy persons was found to be only 5 to 10 per cent and 9 to 14 per cent, respectively, the finding of a high prevalence in *Staph. aureus* and beta hemolytic streptococci on the pillars and gingiva of diabetics is regarded as significant and merits further investigation.

119. Prevalence and Distribution of Extracted First Permanent Molars in Teen-Age Children. *Maury Massler, Leo M. Sreebny, and Harry Goldblatt, College of Dentistry, University of Illinois, Chicago, Ill.* The prevalence and distribution of extracted first permanent molars was determined in 3,570 high school students (1,694 males and 1,876 females) ages 13 to 18 years. One or more first permanent molars had been lost by 26.8 per cent (959) of the children. A total of 1,519 first molars had been extracted in these 1,038 children, or an average of 1.58 first molars per child. Forty-four per cent of the 1,519 extracted first molars were lower left first molars, 34.5 per cent were lower right first molars, 11.5 per cent were upper right first molars, and 10.0 per cent were upper left first molars. There was thus a strong predilection for the loss of lower first molars (78.5 per cent) over upper first molars (21.5 per cent). There was a significantly greater loss of lower *left* over lower *right* first molars but no significant difference in molar loss between males and females and no significant differences in prevalence or distribution of molar loss at any age period between 13 to 18 years.

120. Acid Decalcification of Rat Molar Teeth in Vivo. *Robert F. Muller, Joan F. Thomas, Ann G. Dinse, and Harold C. Hodge, University of Rochester, School of Medicine and Dentistry, Rochester, N. Y.* It has been shown previously that in vivo decalcification of rat molars caused by acid beverages and acid-sugar solutions is much more severe in the lower jaws than in the upper jaws (*Arch. Biochem.* **8**: 405, 1945). Periodic examination of rat molars immersed in acid solutions in vitro revealed qualitatively no apparent difference in enamel loss in the upper and lower molar teeth. To determine whether contact of the solution with the teeth is an important factor in the in vivo difference, adult rats were trained to drink when on their backs; for comparison, litter mates were given the same solutions in the conventional water bottles and permitted to drink in the customary position. Each rat was given 20 ml. of test solution daily. The solutions tested were water (control), a common cola beverage known to contain 0.055 per cent phosphoric acid (pH 2.6), and an aqueous solution of 0.45 per cent lactic acid (85 per cent lactic acid diluted 1:200; pH 2.6). All prepared solutions contained 10 per cent sucrose as a sweetening agent. At the end of one week, the animals were sacrificed and their teeth graded by a visual scoring method (*Science* **102**: 404, 1945). No damage was observed in the control animals (drinking 10 per cent sucrose solutions). The cola-treated rats when "fed" in either position showed moderate enamel destruction in the upper and lower jaws. The lactic acid-treated rats, however, when fed in an upright position showed *maximal* lower molar damage and *slight* upper molar damage confirming the earlier observation; when fed in an inverted position, the damage to both jaws was *maximal.* There is no reason to assume that the upper teeth have a special resistance to or defense mechanism against acid decalcification as compared to the lower teeth. Since far greater time is required for the animals to drink in an inverted position, these data suggest that increased time of contact of the ingested solution with the tooth surface, in vivo, may partially account for the extent of molar decalcification caused by solutions of weak acids.

121. Studies on the pH Tolerance of Oral Bacteria. *Patricia Mullett, Valerie Hurst, and James Nuckolls, University of California College of Dentistry, San Francisco, Calif.* Six strains of oral actinomycetes, five of which produce carieslike lesions in hamster molars in vitro, have been tested for their ability to grow at various hydrogen-ion concentrations. Pour plates were made with nutrient agar adjusted with lactic acid, hydrochloric acid, and sodium hydroxide. In the media adjusted with lactic acid, inhibition occurred in the range between 5.0 and 5.6 and most frequently between 5.2 and 5.6. When

hydrochloric acid was used, inhibition occurred between 4.8 and 5.7 with more frequent values between 4.9 and 5.7. The reason for the greater inhibition by lactic acid is not known. In five strains sodium hydroxide inhibited growth in the range between 9.1 and 9.7 and most frequently between 9.1 and 9.5. The sixth strain showed good growth at a pH of 9.9 with indications of growth at even higher sodium hydroxide concentrations. Preliminary tests on the pH tolerance of five strains of oral lactobacilli indicate that nutrient agar adjusted with hydrochloric acid will support growth at pH 4.0 or lower. In agar adjusted with sodium hydroxide there is little evidence of growth beyond 8.2.

122. Diethylaminoethanol as an Analgetic Agent. *G. W. Rapp, Ph.D., Loyola University School of Dentistry, Chicago, Ill.* Diethylaminoethanol and some related substances can be used as analgetic agents under certain conditions. The drugs are virtually nontoxic and can be given by mouth. Under some conditions these drugs are as effective as aspirin or codeine.

123. A Method for Stabilizing Solutions of Procaine and Allied Drugs. *G. W. Rapp, Ph.D., and B. F. Gurney, M.S., Loyola University School of Dentistry, Chicago, Ill.* Solutions of local anesthetics of the ester type, of which procaine is a good example, can be stabilized by knowing the hydrolysis products and making use of the "common ion effect." In this method soluble ions common to the hydrolysis products can be used to stabilize solutions for a long time. Such solutions are no more toxic than a solution of the anesthetic itself.

124. Lactobacillus Acidophilus Counts and Periodontal Disease. *S. Leonard Rosenthal, D.D.S., and Herbert M. Cobe, Ph.D., School of Dentistry, Temple University, Philadelphia, Pa.* Patients were examined in the order of their appearance: 50 with obvious clinical and roentgenographic signs of periodontal disease were selected, and 50 without such symptoms were chosen as a control group. *Lactobacilus acidophilus* counts were made before institution of treatment. The count of the group with periodontal disease showed a significantly lower level than the control group for individuals of similar age. Patients with gingivitis had higher counts than those with periodontitis.

125. Electron Microscopy of Fibrous Components of Dentin. *David B. Scott, National Institute of Dental Research, and Ralph W. G. Wyckoff, Experimental Biology and Medicine Institute, National Institutes of Health, Bethesda, Md.* It has been demonstrated previously (*Pub. Health Rep.* **62**: 1513, 1947; *J. D. Res.* **27**: 748, 1948) that, in the course of preparing collodion replicas of dentin for electron microscopy, dentinal fibers often withstand the agents used in processing and remain in place on the replicas. By slightly changing the procedures in pre-etching specimens and in the dissolution of the slabs of dentin from the replicas, it has been possible to study further the fibrous components of dentin. Electron micrographs are presented which show the membranous walls of the dentinal fibers, the branching of dentinal fibers, fibers apparently within the dentinal fibers, collagenous fibrils in the matrix and a new type of ultrafine fibers in the matrix.

126. Replacement of Raw Sugar for Refined Sugar in Caries-Producing Rations. *James H. Shaw, School of Dental Medicine, Harvard University, Boston, Mass.* When suitable caries-susceptible strains of cotton rats and white rats were used as experimental animals, the replacement of all the refined sugar in the purified diet (*J. D. Res.* **26**: 47, 1947) by raw sugar did not alter the rates of initiation and development of carious lesions in molar teeth whose

crowns were fully formed at the beginning of the experiment. Studies have not been made to determine whether the above substitution, during periods of tooth development, would alter the susceptibility to dental caries. The rates of growth and well-being of animals fed diets differing only in the source of carbohydrate were identical.

127. Similar Postdevelopmental Dietary Effects Upon the Dental Caries Experience in Caries-Susceptible White Rats and Cotton Rats. *James H. Shaw, School of Dental Medicine, Harvard University, Boston, Mass.* A variety of dietary regimens instituted after formation of the molar crowns was largely completed have been found to result in a lower rate of initiation and development of carious lesions in cotton rats than occurred in littermates fed the control caries-producing ration. Several of these procedures have been tested postdevelopmentally in caries-susceptible white rats to determine if the carious processes in the two species were similarly influenced. In all procedures tested, the influence upon the initiation and development of carious lesions in the white rat were highly similar to the effect already demonstrated in the cotton rat. The following examples have been tested: With greatly restricted caloric intakes, there were large decreases in the initiation and development of carious lesions; when diets composed solely of mineralized whole milk, or of mineralized whole milk to which 10 per cent sugar by weight had been added, or of mineralized evaporated milk, or mineralized evaporated milk to which 10 per cent sugar had been added were fed, essentially no carious lesions developed in a 5-month experimental period; with increases in the fat content, or the protein content of the diet, or of fat and protein simultaneously, at the isocaloric expense of sucrose, there were large decreases in the dental caries experience.

128. The Bactericidal Effect of Essential Oils on Some Bacteria. *H. J. Turkheim, D.M.D., International Serum Institute, London, England.* The following essential oils have been tested for their effect on certain bacteria: Ol. Cassiae, Ol. Cinnam., Ol. Eucalypti, Ol. Melaleuca Alternifolia (Ti-Trol Oil), Ol. Menth. Pip., Ol. Origani, Ol. Thymi, Eucalyptol (cineole), Ol. Caryophyll. (eugenol). Furthermore the following substances derived from essential oils were tested: carvacrol and thymol (saturated solution in almond oil). The following organisms were used: *B. coli, Staph. pyogenes, B. anthrax, B. subtilis, Ps. pyocyanea, S. typhi* (Eberth. Typhosa). Furthermore the author's saliva and a nonhemolytic (j) mouth streptococcus were used. The strains were cultivated on Petri dishes and tested against a round disk of blotting paper (diameter, 10 mm.) soaked in the oil under observation. After 48 hours' incubation at 37° C. the plates were "autographed" by direct printing on to bromide paper in the darkroom. The free zones as result of the bactericidal effect of the oil were measured and compared with a silver standard. This standard had been obtained by the following meethod: disks of blotting paper (diameter 10 mm.) were soaked in ammoniacal silver nitrate (Howe) from which the reducing eugenol had been washed out by chloroform, and were tested against the seven organisms and the author's saliva: the average width of all the free zones, between the edge of the silver blotter disc and the growing culture, for all organism was 2.9 mm. Results: (1) Average effect of each oil against all organism: cineole no bactericidal effect; Ol. Cinnam. 3.9 mm.; eugenol 7.85 mm.; Ti-Trol Oil 8.5 mm.; Ol. Eucalypti 8.6 mm.; Ol. Menth. Pip. 9.3 mm. (average from 2 tests); Ol. Origani 9.8 mm.; Ol. Thymi 10.5 mm.; thymol 11.25 mm.; cassia oil 15.8 mm. (average from 2 tests); carvacrol 19.1 mm. (2) Effect of the oils against each organism and the saliva: *Ps. pyocyanea* proved to be the most resistant, being only slightly influenced by Ol.

Cassiae. *B. coli* was very susceptible to Ol. Menth. Pip. and its growth seemed to be inhibited by the vapor of this oil. Cassia oil had a very strong effect on *B. anthrax*. *B. subtilis*, *S. typhi*, and the streptococcus used were affected most by carvacrol. The effect of the oils against each organism was of the following order: (figures indicate the sum of the free zones caused by the 11 oils): *Ps. pyocyanea*, 4 mm.; saliva, 67.5 mm.; *S. typhi*, 85 mm.; *B. subtilis*, 92 mm.; streptococcus, 92 mm.; *Staph. pyogenes*, 97.5 mm.; *B. anthrax*, 153.7 mm. *B. coli* with 171 mm. proved to be the most vulnerable organism.

129. Tests to Determine Abrasive Qualities of Burnt Coral. *Merrill G. Wheatcroft, Carl A. Schlack, and B. L. Taylor, Dental Division, Naval Medical Research Institute, Bethesda, Md.* Burnt coral is made by placing fresh coral on ignited logs that have been arranged in a pit. This is left approximately one week. The result is a semihard mass with a puttylike core. This soft core is used by the natives of American Samoa as a paint for buildings. The possibilities of using burnt coral as an economic dentifrice has been suggested. A toothbrushing machine was used to test the abrasive quality of the coral. A medium abrasive tooth powder was used as control. Standard, 2 row, nlyon bristle, toothbrushes were used in the machine. Specimens for testing consisted of bilaterally paired teeth and uniform lucite rods. The brushes were adjusted to 75 grams pressure and brushed across the specimens 100,000 times. Drawings of the abraded areas were made by ×30 enlargement silhouette projections before and after running the tests. A planograph was used to measure the projected area destroyed by abrasion. In all tests coral was less abrasive than the tooth powder used as control. In one test lucite rods and distilled water were used to determine the abrasive effect of the brush but no measurements could be made at a x30 enlargement. Coral inhibits growth of *Staphylococcus aureus* No. 209 using the agar plate method. The formation of gas bubbles immediately surrounding the coral could easily be misinterpreted as enhanced bacterial growth. Burnt coral has a pH of 12.5 and is irritating to mucous membranes. The possibility of neutralization by leaching is being investigated.

130. The Organic Elements of the Enamel. V. Histochemical Reactions of the Organic Matter in Undecalcified Enamel. *George B. Wislocki, M.D., Harvard Medical School, and Reidar F. Sognnaes, L.D.S., Ph.D., Harvard School of Dental Medicine, Boston, Mass.* Following appropriate fixations, described elsewhere, ground sections were prepared of extracted teeth of rats, Rhesus monkeys and man. The growing ends of rat incisors were, furthermore, embedded in paraffin and sectioned without decalcification. The individual sections were then stained by various histochemical technics for the demonstration of acid mucopolysaccharides, nucleoproteins, alkaline phosphatase, lipids, and glycogen. The enamel of ground sections of fully mature normal undecalcified enamel was found to be practically inaccessible to the stains employed. However, following fixation in Zenker's fluid, in the preparation for basophilic staining, the enamel proved to be sufficiently demineralized to permit access of the stain to its organic framework. Thus, the prism sheaths, when not accidentally lost, were found to stain with basic dyes, such as methylene blue. This staining occurred at a pH as low as 3, a result indicating that the sheaths contain a relatively strong acid substance. Furthermore, after fixation in basic lead acetate and staining with toluidine blue, the prism sheaths exhibited metachromasia, visible particularly in the relatively less calcified sites of the tufts and Schreger's bands. The combination of these two histochemical reactions suggests the presence of an acid mucopolysaccharide in the organic framework of the enamel. In the initial enamel formation

where calcification is barely beginning, a greenish coloration has been found after toluidine staining. This is followed by the reddish metachromasia which, occasionally, may persist in the lesser calcified areas of erupted tooth enamel. Alkaline phosphatase could not be demonstrated in the enamel except in that immature portion described above as staining green with toluidine blue. Sudan black bacillus stain gave a delicate sudanophilic reaction in the developing enamel of growing rat incisors. The periodic acid Schiff procedure did not suggest the presence of glycogen in the organic framework of the enamel.

131. Patterns of Craniodental Orientation. *Maurice R. Zingeser, D.D.S., Tufts College Dental School, Boston, Mass.* Two sets of observations concerning patterns of orientation manifested by the maxillary first permanent molar in its special relationship to the cranial base area and to adjacent maxillary and mandibular dental units are described. The Margolis cephalostat was used to obtain standardized lateral cephalic tracings. All individuals were white, characterized by the presence of a full complement of teeth commensurate with developmental level, and had a minimum of dental crowding. Facial pattern was not a factor in the selection of this material. The study was confined to Class I occlusal normals and untreated Class II, Division I cases. The observation that the maxillary first permanent molar reference point and the incisal edge of the permanent first incisor lie at approximately the same distance from nasion was confirmed statistically. No statistically valid difference was found to exist in this respect between the Class I series and the Class II, Division I group. The second pattern consists of a complex geometric expression of the position of the maxillary first permanent molar reference point in terms of a phase of the spacial configuration of three cranial base points. The angular properties of this geometric construction are such as to suggest the existence of a widespread common pattern of craniodental orientation for this tooth in the age group studied, within the framework of individual anatomical differences. The pattern is equally applicable to both the pre-eruptive and posteruptive positions of this tooth. No valid statistical difference between the Class I and Class II, Division I groups was found to exist in respect to this pattern. Finally, a relationship was found to exist between the construction and the sagittal orientation of the incisal edge of the permanent mandibular central incisor.

XII. Executive Proceedings and Register of Newly Elected Officers and Members

A. COUNCIL SESSIONS

Activities Between General Meetings

Approved the selection of Chicago, the Drake Hotel, and June 24-26, 1949, as the place and time of the Twenty-seventh General Meeting. Nominated the Council candidates for 1949-1950 officers to be listed on the official ballot form. Approved the distribution of disbursements for the Secretary-treasurer's budget.

First Session, Friday, 4:30 P.M., June 24, 1949

President Allan G. Brodie called the meeting to order and announced the attendance of a quorum. The financial report of the Secretary-treasurer was called for.

Receipts for the 1948 Calendar Year

From dues and subscriptions to *J. D. Res.*	$2,638.00
From The C. V. Mosby Co., royalty	1,800.00
From bond interest	32.50
Total	$4,470.50

Disbursements for 1948

To The C. V. Mosby Co., on *J. D. Res.* account	$2,673.93	
For officers' expenses	983.42	
Printing, programs, ballots, etc.	144.00	
Total		$3,801.35
Excess of receipts over disbursements		$ 669.15

Detail of Disbursements from The C. V. Mosby Company

To H. B. G. Robinson, expenses of Editor's office	$ 500.00	
To J. Frank Hall, expenses of Bus. Mgr's office	250.00	
To The C. V. Mosby Co., for extra *J. D. Res.* pages, 1948	831.93	
Total		$1,581.93
Balance unexpended from 1949 royalty		$ 218.07

Status of William J. Gies Special I.A.D.R. Endowment Fund

Bonds (U. S.) on hand	$1,300.00	
Savings bank account balance	221.06	
Total		$1,521.06

Status of General Fund (From $1.00 of Each Member's Dues)

Balance on hand from excess of receipts over expenditures	$ 85.92

On Hand as of June 1, 1949

In savings account No. 8583, State Bank and Trust Co., Evanston, Ill.	$ 221.06
In savings account No. 13548, State Bank and Trust Co., Evanston, Ill.	1,022.20
In checking account, State Bank and Trust Co., Evanston, Ill.	1,837.29
Total on deposit	$3,080.55
June 1, 1948, Total on deposit	$3,291.22

On motion this report was approved and filed for printing in the official proceedings. It was moved and seconded that an over-all budget for the 1949 calendar year be approved as follows:

For expenses of the Secretary-treasurer's office	$ 700.00
For expenses of Editor and Business Manager, including extra pages in the 1949 *J. D. Res.*	$1,800.00
For the block member subscriptions to the *J. D. Res.* at the rate of $4.00 per subscribing member	

A communication from the New York Academy of Dentistry, signed by its Secretary, Edward C. Stillwell, was read. This contained three proposals, namely, that:

"1. A gift of $10,000, payable at $2,000 annually to begin in December, 1949, be accepted. This amount is to be used in support of the current publication of the *Journal,* to replace the annual payment to the ASSOCIATION by The C. V. Mosby Company of the $1,800, under the current contract.

"2. The front cover (of the *J. D. Res.*) carry the statement, 'Published by the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH, with the cooperation of The C. V. Mosby Company, etc.'

"3. The New York Academy of Dentistry has been assured that it will receive the gift of $10,000, to be paid to the ASSOCIATION—by the Academy as the formal donor—in annual installments of $2,000 as specified in paragraph 1, above. Dr. Gies has conducted the financial negotiations to this end and, at any time before December, 1949, will *as a member of the Academy* present to the Academy War Savings Bonds for the full amount, to assure these payments by the Academy to the ASSOCIATION (I.A.D.R.) as stated for the purposes indicated."

Copies of these proposals, the conveying letter, and the adjustments that would be required in the current publishing agreement were supplied to members of the Council. After a long and full discussion covering each item of the three proposals and balancing the advantages and disadvantages of accepting the $10,000 under the terms both stated and implied in the three proposals, it was formally moved and seconded that the proposals of the New York Academy of Dentistry and the associated gift of $10,000 be rejected and that a committee of three be appointed to draft a suitable reply to the Academy. This was carried by a vote of 24 to 0 (Council attendance at this session was 24). The chair appointed B. G. Bibby, J. R. Blayney, and P. C. Kitchen as a committee to draft the reply and report back to the Council at the Sunday morning session.

Fifty-one nominations to membership were submitted to the Council for disposal. A committee of four was authorized to study these and report back their findings to the Council at the Sunday session. Committee: Armstrong, Blayney, Hatton, and Schour.

A resolution having to do with current legislative proposals with respect to the practice of medicine and dentistry was submitted, discussed, and no action taken. The activities and objectives of the National Society for Medical Research were heartily endorsed and the ASSOCIATION, including individual members, were urged and authorized to cooperate in every way possible. A communication from the Children's Bureau with reference to its Clearing-house for Research in Child Life was read for the information of ASSOCIATION members and the work approved.

Some discussion concerned the time and place of the next general meeting. A preference poll taken at the close of the discussion disclosed that of those present, 14 favored a March time, 8 June, and 2 no preference.

Council adjourned till Sunday morning.

Second Session, Sunday, 8:30 A.M., June 26, 1949

President Brodie called the Council to order with a full quorum and called for the report of the committee appointed to draft a reply to the communication from the New York Academy of Dentistry. It follows:

"To the New York Academy of Dentistry:

"At the annual meeting of the INTERNATIONAL ASSOCIATION OF DENTAL RESEARCH held in Chicago, June 24-26, 1949, the Council of the ASSOCIATION gave full and careful consideration to your letter of May 27, 1949.

"It was decided that the suggestions contained in that letter regarding the management of the *Journal of Dental Research* do not offer any advantages over the present arrangement. Therefore the Council voted unanimously not to accept the suggestions contained in your letter. Furthermore it was the definite feeling of the Council that the *Journal* in its present form and mode of publication meets all the requirements of a first class, ethical publication and that it is serving admirably its purpose in the promotion of dental research. It was the decision of the Council that the statement on the cover reflects accurately the pub lication arrangements.

"Despite this negative action the Council wishes the Academy to know that it appreciates the Academy's interest in the welfare of the *Journal* and that it will strive continuously to maintain the *Journal* as a publication of which we can all be proud."

After some discussion and in the absence of suggestions for any modification of the committee's draft, on motion the Secretary-treasurer was ordered to transmit a copy each to the Secretary of the New York Academy of Dentistry and to William J. Gies, both copies to be signed by him as Secretary-treasurer of this ASSOCIATION. Vote poll: Yes, 22, No. 0; all Councillors except the Secretary-treasurer voting.

Councillor Jay moved that a committee be formed to advise and support the program committee in efforts to construct better programs for the annual meetings. Seconded and carried. By motion the ASSOCIATION was advised to fix the dues for the 1949-1950 period at the rate of $5.00 per member per each 12 month period through Dec. 31, 1950, and limited to members residing in North America. Of each $5.00 item $1.00 is to be appropriated for the general fund as budgeted for use by the Secretary-treasurer and $4.00 for member block subscriptions to the *Journal of Dental Research.* (Nominal dues for members residing outside of North America remain the rate of $1.00 per year, with subscription to the *Journal* optional.) On motion disbursements were authorized, if approved by the ASSOCIATION at the Sunday executive session, from The C. V. Mosby $1,800 royalty fund, for both the expenses of the offices of the Publication Committee and for the purpose of increasing the number of pages in the 1949 volume of the *Journal of Dental Research* above the stipulated 600 pages per volume at the discretion of the Editor. Approved the nominations of 38 candidates for membership for submission to the ASSOCIATION for election to membership at the final executive session. Approved the formation of two new sections, at Los Angeles and in Nebraska, respectively. Authorized the formation of a committee to study the publicity problems associated with annual meetings and especially releases to the press of program content and articles submitted for publication in the *Journal of Dental Research.* There being no more business, the Council adjourned.

Councillors present at one or both of the Council sessions: Arnold, Agnew, Barr, Bibby, Blayney, Boling, Boyle, Brashear, Brodie, Bryan, Burrill, Costich, Crawford, Dean, Dobbs, Fleming, Hatton, Hill, Hine, Jay, Kitchen, Neuwirth, Noyes, Peterson, Peyton, Robinson, Swanson, Zander. Total, 28.

B. REPORT OF THE BOARD OF EDITORS 1948-1949 (VOLUME 27)

The first volume of the *Journal* published under the new contract was completed in December, 1948. From the viewpoint of the Editor and Business Manager this arrangement was entirely successful and they wish to compliment the publishers, The C. V. Mosby Company, for their excellent cooperation.

Volume 27 included 805 pages. These were made up by 85 original papers, 6 Editor's Viewpoints, 5 Section Proceedings, the Proceedings of the 26th General Meeting of the INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH and miscellaneous matter, including the index. Although our contract calls for only 600 pages per volume, the additional pages were made available by purchase of $831.93 in extra pages from the money received as a royalty from the printer and by agreement that the cost of the 209 pages of the Proceedings of the Michigan Dental Caries Conference should be shared by the publisher. We believe that the format, paper, illustrative material, and press work are to the credit of the publisher and the ASSOCIATION which own the *Journal.*

The activity in dental research and interest of authors in publishing in the *Journal of Dental Research* appear to be increasing. Despite the increased size of our *Journal* we now have over 70 acceptable papers on hand. We have purchased 36 extra pages in the June issue of the *Journal* at a cost of $500.00. This will allow the publication of approximately 4 additional papers. We plan on purchasing 72 more pages in the December issue.

The Editor expressed his thanks to Doctors Paul C. Kitchin and J. B. Brown, assistants to the Editor, for their invaluable assistance and to the many members of the ASSOCIATION who have served as referees and readers for various special papers. He also thanked Doctor E. H. Hatton whose assistance makes the publication of the ASSOCIATION'S Proceedings possible.

The Business Manager presented the following report:

REPORT OF THE BUSINESS MANAGER OF
JOURNAL OF DENTAL RESEARCH

From January 1, 1948 to December 31, 1948

Balance on hand December 31, 1947		$6,392.37
Receipts:		
Back issues of the *Journal*		683.65
		$7,076.02
Disbursements:		
Clerical & Misc.	$ 568.58	
Printing	5,091.17*	
Stationery & Supplies	143.97	
Postage	188.69	
Total		$5,992.41
Balance on hand		$1,083.61

*Balance from Volume 26.

J. FRANK HALL
Business Manager

The Business Manager further reported that The C. V. Mosby Company had presented the Publication Committee with a copy of their expense sheet for Volume 27. This showed expenses of $17,047.98 and revenue of $10,742.50, with a deficit of $6,305.48 which has been assumed by The C. V. Mosby Company.

HAMILTON B. G. ROBINSON
Editor

WILLIAM J. GIES ENDOWMENT FUND, JOURNAL OF DENTAL RESEARCH

Financial Statement as of June 24, 1949

Compiled by LEUMAN M. WAUGH, D.D.S., Treasurer

This Fund was created, on the initiative of the voluntary Committee on Endowment—organized in New York City in 1937—to insure the continuance of the *Journal of Dental Research* "in full accord with the highest ideals of strictly professional journalism devoted to the advancement of research," to which the *Journal* was dedicated at its establishment in 1919.

I. *Amounts Received From Organizations*
(June 20, 1948, to June 24, 1949)

Louisiana State Dental Society	$ 25.00	
Ohio State Dental Association	120.00	
		$145.00

II. *Amounts Received From Individuals*
(June 20, 1948, to June 24, 1949)

Total contributions 1948-1949 ---------- $145.00

III. *Interest, Income and Expense (current year)*

No expenses were recorded.

INTEREST TO DATE, JUNE 24, 1949, ON WAR BONDS:

Number of Bonds	Denomination of Bonds	Date Purchased	Interest Accrued During Current Year
10	$1,000.00	March 1942	$ 260.00
22	1,000.00	May 1942	572.00
8	1,000.00	July 1942	208.00
2	1,000.00	Dec. 1942	50.00
1	1,000.00	March 1943	25.00
1	25.00	Feb. 1944	.52
5	1,000.00	March 1944	105.00
2	1,000.00	May 1944	42.00
1	1,000.00	July 1944	19.00
3	1,000.00	Sept. 1945	39.00
2	1,000.00	March 1946	22.00
Total			$ 1,342.52

Total contributions 1948-1949 (Section I and Section II) --- 145.00

Total addition to the Fund, including accrued interest for the current year ---------- $ 1,487.52

IV. *Summary of the Total Assets: 1937-1949*

Total receipts (assets) for the Fund as of June 24, 1949		$46,345.73
Paid for 56 War Bonds (Section VI)	$41,440.00	
Shares of stock (Section VI)	365.00	
Interest accrued during current year	1,342.52	
Interest accrued during 1946-1948	2,316.69	
Balance	881.52	

V. *Cumulative Summary of Annually Recorded Items of Receipts and Expenditures*

Year	Contributions (Payments)	Earned Interest	Expense	Net Receipts	Cumulative Annual Net Receipts
1937-38	$10,652.50	None	$134.06	$10,518.44	$10,518.44
1938-39	4,748.17	None	None	4,748.17	15,266.61
1939-40	5,693.78	None	None	5,693.78	20,960.39
1940-41	5,190.55	$ 206.78	3.53	5,393.80	26,354.19
1941-42	4,166.35	333.95	None	4,500.30	30,854.49
1942-43	1,766.31	112.19	None	1,878.50	32,732.99
1943-44	3,302.23	334.00	None	3,636.23	36,369.22
1944-45	3,744.45	459.05	None	4,203.50	40,572.72
1945-46	1,144.50	497.17	None	1,641.67	42,214.39
1946-47	249.50	1,183.29	None	1,432.79	43,647.18
1947-48	77.63	1,133.40	None	1,211.03	44,858.21
1948-49	145.00	1,342.52	None	1,487.73	46,345.73
	$40,880.97	$5,602.35	$137.59	$46,345.73	

VI. *Statement Regarding Investment in War Bonds*

By authorization of the Trustees of the ASSOCIATION, fifty-six War Bonds of Series F ("12-year appreciation bonds"), costing $740 each, have been

purchased from the funds in the Endowment Committee's Treasury, as of the following dates and serial numbers:

Purchased (10) March	1942:	M140400F to M140409F, incl. --	$ 7,400.00
Purchased (22) May	1942:	M197811F to M197832F, incl. --	16,280.00
Purchased (8) July	1942:	M237558F to M237565F, incl. --	5,920.00
Purchased (2) December	1942:	M423990F and M423991F -----	1,480.00
Purchased (1) March	1943:	M204356F ----------	740.00
Purchased (5) March	1944:	M750832F to M750836F, incl. --	3,700.00
Purchased (2) May	1944:	M752375F and M752376F -----	1,480.00
Purchased (1) July	1944:	M892919F ----------	740.00
Purchased (3) September	1945:	M649868F to M649870F, incl. --	2,220.00
Purchased (2) March	1946:	M1061926F and M1061927F ---	1,480.00
			$41,440.00

The payee named on each bond is "Committee on the William J. Gies Endowment Fund for the *Journal of Dental Research* (an unincorporated body)." All of these War Bonds are in the custody of the treasurer of the Committee, in a safety deposit box, Empire Trust Company, 580 Fifth Avenue, New York 19, N. Y., in trust for the Committee.

The contribution of $18.50, listed in the Section II of the report for 1943-1944 from Dr. Joseph E. Psayala, was in the form of a $25 United States War Bond Series F ("12-year appreciation bond"), Q631670F, the payee being "William John Gies Endowment Fund for the *Journal of Dental Research.*" It is in the custody of the Treasurer in a safety deposit box, Empire Trust Company, 580 Fifth Avenue, New York 19, N. Y., in trust for the Committee. It was purchased in February, 1944.

The Farnsworth Television and Radio Corporation Stock, indicated in Section IV, is also in the custody of the Treasurer of the Committee in a safety deposit box, Empire Trust Company, 580 Fifth Avenue, New York 19, N. Y., in trust for the Committee.

VII. *Summary of Open Accounts Relating to Pledges*

	Pledge	Paid	Balance	
A. Organizations				
Kansas: First District Dental Society ----------	$ 100.00	$ 25.00	$ 75.00	
J. B. Mann Study Club, Washington, D. C. ----	1,000.00	200.00	800.00	
				$ 875.00
B. Individuals				
Harry Kaplan ----------	250.00	100.00	150.00	
R. J. Rinehart ----------	200.00	75.00	125.00	
				$ 275.00
				$1,150.00

Respectfully submitted,
LEUMAN M. WAUGH, Treasurer

C. EXECUTIVE SESSIONS

First Session, Saturday, 9:15 A.M., June 25, 1949

The meeting was called to order by the President, Allan G. Brodie, with a brief welcome to members and visitors repeating essentially his remarks at the opening of the first scientific session of Friday. The minutes of the twenty-sixth General Meeting were offered as printed in the December, 1948, issue of the *Journal of Dental Research.* Since no corrections were submitted and no call heard for reading of the minutes, the minutes were ordered approved as printed in the proceedings of the 1948 General Meeting. Harold J. Leonard, J. L. T. Appleton and L. S. Van Kirk were appointed to receive and count the ballots for the 1949-1950 general officers. The Secretary-treasurer reported the loss of the following members: (A) By resignation: Col. H. E. Ash, M. L. Deakins, Wm. E. Ehrich, L. L. Eisenbrandt, H. T. Fraser, J. Raymond Gill, Geo. W. Grieve, Alexander Ker, Kenneth W. Ray, John B. Williams. (B) By death: István Bánhegyi, Béla Bonyhárd, János Brand, Károly Ferenczy, Oszkár Kömives, Ferenc Köszeg, Victor C. Myers, Rudolph Rehák, Henrik Salamon, Harry M. Semans, Lászlo Sugár, Dénes von Mathe, Harold L. Weatherford, Daniel E. Ziskin. At the request of the President, members stood briefly as a token of respect to the memory of their former colleagues and in appreciation of their contributions to the promotion of research. Session adjourned.

Second Session, Saturday, 3:30 P.M., June 25, 1949

The ASSOCIATION was called into executive session to listen to the report of the Treasurer of the William J. Gies Endowment Fund for the *Journal of Dental Research,* which, in the absence of Dr. L. M. Waugh, was read by the Secretary-treasurer. This report was accepted and ordered to be printed in the proceedings of this meeting. The Treasurer and the Committee were voted hearty thanks for their services in this cause. Session adjourned.

Third and Final Session, Sunday, 3:30 P.M., JUNE 26, 1949

With President Brodie presiding, the report of the tellers was received and the results of the election announced. By motion all recommendations and acts of the Council were approved and authorized as recorded in the Council report presented by the Secretary-treasurer. Included with other actions are, specifically: the annual dues assessment of $5.00 per North American member; authorization of disbursements for the support of the offices of the Secretary-treasurer and the Publication Committee, including the block membership subscriptions to the *Journal of Dental Research;* and the Council action with respect to the communication from the New York Academy of Dentistry.

On motion of the Secretary-treasurer, carried without a dissenting vote, the rules were suspended and the Secretary-treasurer directed to cast a unanimous vote for the election to membership of all persons on the list previously approved by the Council for such action and read as a part of the motion. (See page 687 for this list.)

The Council was authorized to act for the ASSOCIATION in the interval between this and the next general meeting, and especially to select the time and place of the next meeting. A hand poll of the members in attendance indicated a slight preference for a June time rather than March. The Secretary-treasurer was authorized and directed to convey the appreciation of the ASSOCIATION to the many persons and groups who were instrumental in cre-

ating the setting for one of the best, if not the best, general meeting in the history of the ASSOCIATION. Especially cited were the staff and Mr. Dick Flynn of the Drake Hotel, Dr. Weinmann and the group from the University of Illinois supplying the projectors and the projection service, and the girls at the registration desk from Dr. Fosdick's department at Northwestern University.

Dr. Brodie then presented the new officers and duly installed them in a brief but impressive ceremony. An unscheduled feature of this portion of the proceedings, deeply appreciated by the Secretary-treasurer, were the remarks and presentations from officers and members for his services for the ASSOCIATION.

The last item on the program of the meeting was the address of the incoming president, J. Roy Blayney, who was suitably introduced by Dr. Brodie with reference to his noteworthy achievements and the honor he has conferred on this ASSOCIATION in accepting the office and responsibility of its presidency. At the close Dr. Blayney received the gavel, the symbol of his new office, and after calling for any new business formally adjourned the 1949 General Meeting.

D. OFFICERS AND MEMBERS ELECTED

ACTIVE OFFICERS: President—*J. Roy Blayney,* Zoller Memorial Dental Clinic, University of Chicago. President-elect—*Basil G. Bibby,* Eastman Dental Infirmary. Vice-president—*Leonard S. Fosdick,* Northwestern University. Secretary-treasurer—*Edward H. Hatton,* Northwestern University. Trustee (for a five-year term)—*Harold C. Hodge,* University of Rochester.

MEMBERS ELECTED: Berkeley, Calif.—*C. A. Ostrom,* Naval Medical Research Unit No. 1. Boston, Mass.—*Robert L. Glass,* Tufts College and *Kalevi Koski,* Forstyth Dental Infirmary. Brooklyn, N. Y.—*Albert E. Sobel,* Jewish Hospital of Brooklyn. Bethesda, Md.—*Clarence E. Dawson* and *James A. English,* Naval Medical Research Unit. Bogota, Colombia—*Alberto G. Barriga.* Chicago, Ill.—*Stewart A. Koser,* University of Chicago and *Alexis W. Maier,* University of Illinois. Cleveland, Ohio—*Robert J. Bruckner, Sholom Pearlman, Frank E. Walter,* and *Betty Jane White,* Western Reserve University. Columbus, Ohio—*John J. Ennever,* Ohio State University. Corvallis, Ore.—*Demetrios M. Hadjimarkos,* Oregon State College. Helsinki, Finland—*Juuso J. T. Klimäki,* University of Helsinki. Great Lakes, Ill.—*W. E. Ludwick,* Naval Training Corps. Iowa City, Iowa—*Kenneth E. Wessels,* State University of Iowa. Kansas City, Mo.—*Norman A. Moore, Hugh I. Myers,* and *Benjamin Warner,* University of Kansas City. Lincoln, Neb.—*Donald T. Weggener,* University of Nebraska. Los Angeles, Calif.—*Lucien A. Bavetta, Sol Bernick,* and *Robert L. Rutherford,* University of Southern California. Medica, Pa.—*Herbert M. Cobe,* Temple University. New York City—*Earl O. Butcher,* New York University, *David Dragiff,* Columbia University, *Eli Goldsmith,* New York University, and *Leon Lieber.* Philadelphia, Pa.—*I. B. Bender* and *Samuel Seltzer,* University of Pensylvania. Pittsburgh, Pa.—*Kenneth H. Strader,* Lee S. Smith & Son. Portland, Ore.—*Frank G. Everett* and *N. M. Phatak,* University of Oregon. San Francisco, Calif.—*Harold G. Ray,* University of California. Topeka, Kan.—*Leon R. Kramer,* University of Kansas City. Toronto, Canada—*John B. MacDonald,* University of Toronto. Total, 38.

E. MEMBERS VOTING AT THE ANNUAL ELECTION

Aisenberg, Allen, Anderson (G. M.), Arnim, Arnold, Artagaveytia, Austin, Bagnall, Barr, Bauer, Bear, Becks, Bernier, Beube, Bibby, Blackberg, Blackwell, Blayney, Blumenthal, Bodecker, Boucher, Bradley, Brand, Brandhorst, Brashear, Breitner, Brown (J. B.), Brown (J. C.), Brucker, Brudevold, Burkett, Burrill, Canby, Cheyne, Chilton, Clark, Clausen, Clawson, Clough, Costich, Coolidge (T. B.), Cowling, Cox, Coy, Crawford, Cross, Crowley Dahlberg, Dale, Darlington, Davis, Dean (H. T.), Deatherage, Dickson, Dobbs, Dreizen, Dunning (J. B.), Dummett, East, Easlik, Edwards (L. F.), Edwards (R. W.), Elliot (F. C.), Ellis (R. G.), Epstein (I. A.), Erikson, Fairbank, Fastlicht, Ferguson, Figg, Finn, Fisher, Fleming, Fosdick, Fox, Freeman (C. W.), Frey, Gabel, Gafafer, Gamboa, Gilda, Gilkison, Ginn, Glickman, Godwin, Goldberg (H. J. V.), Goldman, Goodsell, Granados, Grossman, Gurley, Hadley, Hahn, Hill, Hine, Hodge, Hoffman, Hooper, Hubbell, Hughes (B. O.), Irving, Jarabak, Jasper, James, Jordan, Kaiser, Kaletsky, Keilty, Kerr, Kesel, Keyes, Khalifah, King, Kitchin, Klaffenbach, Kniesner, Knighton, Koch, Krasnow, Krogh, Kurth, Lalonde, Land, Lazansky, Leicester, Levy, Lieban, Losch, Lovestedt, Lowery, Lundquist, Main, Mann, Manly, Margolis, Marjerison, Marshall-Day, Martin, Massler, McCauley, McClure, McCollum, McCrea, Merkeley, Merritt, Mercado, Milhon, Millberry, Mine, Mitchell,

Moore (G. R.), Moore (Mary), Moore (T. E.), Morse (Anna), Mueller (A. H.), Mueller (Emil), Myers, Nelson, Neuwirth, Nizel, Noyes (H. J.), Nuckolls, Oblatt, Oliver (O. A.), Orban, Orland, Paffenbarger, Parke, Patterson, Permar, Peterson (H. W.), Peyton, Pheiffer, Phillips, Pincus, Rabkin, Radusch, Rault, Revesz, Reynolds, Ritchie, Robinson (H. B. G.), Romnes, Rosenstein, Rudolph, Ryder, Salzman, Sartori, Scherer, Schlack, Schour, Scott (D. B.), Scrivener, Shaner, Seidler, Shaw, Shourie, Sicher, Simon, Simmonds, Sloman, Smith, Smyd, Snyder, Sognnaes, Sorrin, Stewart, Stout, Swanson (H. A.), Swanson (W. F.), Sweet, Teuscher, Van Kirk, Volker, Volland, Wach, Wainwright, Waldo, Waugh, Wheeler, Wilbur, Willett, Williams, Willman, Worman, Wright, Wylie, Zander. Total, 229.

F. MEMBERS IN ATTENDANCE AT ONE OR MORE SESSIONS, LISTED BY SECTIONS

Ann Arbor—Easlik, Jay, Kerr, Peyton, Sweeney. *Baltimore*—Aisenberg, Dobbs, Hahn, McCrea, Robinson. *Boston*—Brudevold, Cohen, Dunning, Glickman, Greep, Hatch, Keyes, Lazansky, Manly, Marshall-Day, Nizel, Sognnaes, Waldo, Zander. *Chicago*—Blackwell, Blayney, Bradel, Brodie, Calandra, E. D. Coolidge, T. B. Coolidge, Cooper, Dahlberg, Denton, Engel, Fosdick, C. W. Freeman, Gruebbel, Harrison, Hatton, Jarabak, Kesel, Krashen, Kurth, Martin, Massler, Orland, Rapp, Shell, Sarnat, Schour, Skillen, Skinner, Steinmeyer, Thompson, Tilden, Tylman, Wallace, Wassermann, Weinmann. *Cleveland*—Burwasser, S. W. Chase, Hill, Kniesner. *Columbus*—Edwards, Kitchin, Permar, Robinson. *Houston*—Barr, Elliott, Telford. *Indiana*—Hall, Hine, Muhler, Phillips, Swenson, Van Huysen. *Iowa City*—Boyd, Bryan, Easton, Higley, Kern, Klaffenbach, Smith. *Louisville*—Burrill, Wilbur. *Minnesota*—Armstrong, Clark, Crawford, Lovestedt, Mitchell, Simon, Speidel. *New Orleans*—Peterson. *New York*—East, Leonard, Neuwirth, Wright. *Oregon-Washington*—Brauer, Cheyne, Jump, Law, Noyes, Snyder, Thomas. *Philadelphia*—Appleton, Boyle, Crowell, Gabel, Rosenthal, Taylor, Williams, Winter. *Pittsburgh*—Cox, Swanson, Van Kirk. *Richmond*—Bear, Brashear, Clough. *Rochester*—Bibby, Costich, Dale, Hodge, Shourie, Simmons. *St. Louis*—Bauer, Boling, Brandhorst, Gurley, Knighton, Levy, Main. *San Francisco*—Agnew, Becks, Fleming, Leicester, Ryder, Sloman, Wylie. *Tennessee*—Ginn. *Toronto*—R. G. Ellis. *Washington*—Arnold, Bernier, Caul, Dean, Hampp, Losee, McCauley, McClure, Oartel, Paffenbarger, Schoonover, Scott, Stephan, Zipkin. *No Section Membership*—Bradley, Brumfield, Dreizen, Frey, Finn, Hooper, Ireland, Keys, Mann, Molnar, T. E. Moore, Nelson, Scrivener, Smyd, Toverud, Volker, Wessinger, Wise. Total, 173.

Attendance of New Members Elected June 26, 1949—J. A. English, John Ennever, F. G. Everett, R. L. Glass, S. A. Koser, W. E. Ludwick, A. W. Maier, N. A. Moore, H. I. Myers, Sholom Pearlman, N. M. Phatak, H. G. Ray, B. W. Warner, K. E. Wessels, Betty J. White. Total, 15. Registered Guests, 214.

Total attendance, 402.

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[1]Abbreviations used: I.A.D.R.—International Association for Dental Research; J.D.R.—*Journal of Dental Research*; A.D.A.—American Dental Association.
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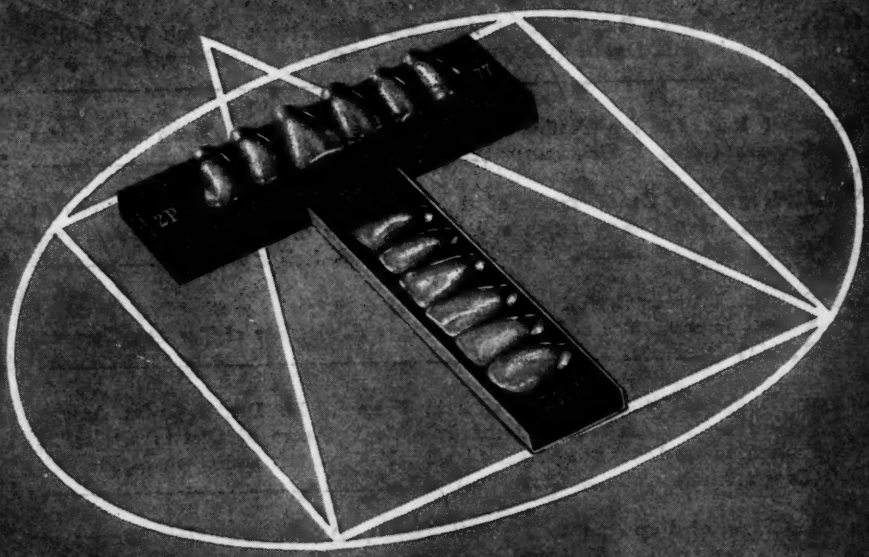

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